First Author: Joseph-Patrick Clarke (Postdoctoral)	Poster Session: pm
Presenting Author: Joseph-Patrick Clarke (Postdoctoral)	Location: 1
Mentor/Lab: Christopher Donnelly	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	

Title: Inducing Stress Granule Formation Using Optogenetics

Summary: The goal of this work is to generate light-induced SGs to study the role of these membraneless organelles in ALS/FTD. Our work is the first to report the formation of functional membraneless organelles using light and demonstrates spatial and temporal control in their formation in the absence of cytotoxic cell stress. Employing this method allows us to broaden our understanding of the pathobiology underlying ALS and FTD and their neuropathologies.

Abstract: Amyotrophic lateral sclerosis (ALS) and Frontotemporal dementia (FTD) are progressive fatal neurological diseases caused by the loss of upper and lower motor neurons or cortical neurons respectively. The majority of diagnosed ALS and FTD patients are classified as having a sporadic phenotype with the remaining considered familial based on patient history. A molecular similarity between both neurological diseases is the observed cytoplasmic aggregation of the RNA-binding proteins TDP-43 and FUS in post-mortem tissue samples. Current hypotheses suggest that impaired homeostasis of cell stress activated cytoplasmic granules called stress granules (SGs) may serve as sites of TDP-43 and/or FUS aggregation in disease and thus may promote disease progression. SGs form under periods of cell stress and function to prevent global protein synthesis to promote the upregulation of stress response genes until the stress is removed. Elucidating such an effect however has been problematic using current methods to form stress granules since prolonged treatment with extracellular stress is cytotoxic thus preventing the study of prolonged or repetitive stress granule formation on in the induction of ALS/FTD neuropathology. To overcome this we developed a novel method employing light-induced protein clustering to seed the core protein components. The goal of this work is to generate light-induced SGs to study the role of these membraneless organelles in the absence of toxic extracellular stressors. Employing this method we are able to broaden our understanding of the pathobiology underlying ALS and FTD and their neuropathologies. Our results demonstrate that the light induced SGs co-localize with endogenous stress granule components including G3BP1 Ataxin-2 PABPC1 TIAR and eIF3H. Additionally the light-induced SGs sequester mRNAs and translation factors to inhibit global protein synthesis similar to endogenous SGs. Light-induced SGs can be controlled to induce prolonged or repetitive SG formation and light-induced SGs sequester with the ALS/FTD proteins TDP-43 and FUS. This body of work allows us to form functional SGs with great spatial and temporal control and in the absence of cytotoxic cell stress. This is the first report of the formation of functional membraneless organelles using light. We are currently using this tool to elucidate the role of SGs in TDP-43 and FUS aggregation and in ALS/FTD pathobiology.

First Author: Amanda Gleixner (Postdoctoral)	Poster Session: pm
Presenting Author: Amanda Gleixner (Postdoctoral)	Location: 2
Mentor/Lab: Christopher Donnelly	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	

Title: Evaluation of FG Nup deficits in C9ORF72 ALS

Summary: Proper cellular function relies on the transport of molecules between the nuclear and cytoplasmic compartments. However deficits in nucleocytoplasmic trafficking have been observed in C9ORF72-associated ALS but why this occurs in the disease remains unknown. This work exams defects in the protein responsible for nucleocytoplasmic transport the nuclear pore complex and seeks to rescue neuronal dysfunction by reversing nuclear pore complex deficits.

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive and fatal neurodegenerative disease characterized by the degeneration of the motor neurons and interneurons in the brain and spinal cord. Ninety percent of ALS cases occur sporadically (sALS) while approximately 10% of cases are linked to genetic mutations which are associated with a family history of the disease (familial ALS) (Renton et al. 2013; DeJesus-Hernandez et al. 2013). The most common genetic mutation associated with both sporadic and familial ALS is attributed to a GGGGCC hexanucleotide repeat expansion (HRE) in the first intron of the C9orf72 gene. Patients with C9ORF72 ALS may show up to hundreds or thousands of GGGGCC repeats while fewer than twenty repeats are typically observed in control patients (DeJesus-Hernandez et al. 2012). Neurotoxicity of the GGGGCC HRE in C9ORF72 ALS has been associated with the generation of toxic GGGGCC RNAs and dipeptide repeat proteins (DPRs) that are synthesized from the GGGGCC HRE by the non-canonical repeat associated non-ATG translation (RANT) pathway (Donnelly et al 2013; Wen et al 2014; Ash et al 2013). Recent studies have shown that nucleocytoplasmic transport pathways are greatly perturbed by the GGGGCC HRE (Zhang et al 2015; Freidbaum et al 2015; Jovičić A). Nucleocytoplasmic transport is driven by the nuclear pore complex (NPC). The NPC is comprised of approximately 30 different proteins that are termed nucleoporins. Nucleoporins have been shown to be modifiers of both nuclear transport defects and neurodegeneration in C9orf72 ALS Drosophila models (Freidbaum et al 2015; Boeynaems et al 2016). The permeability and selectivity barrier of the NPC is comprised in part by a class of nucleoporins with phenylalanine-glycine repeat domains (FG Nups). We performed a comprehensive analysis of FG Nups in cellular Drosophila models of C9orf72 ALS and are validating these findings in ALS patients tissue. We observed that modulation of various FG Nup levels altered neurotoxicity in C9orf72 ALS Drosophila models. Furthermore FG Nup deficits were detected in C9ORF72 ALS cellular models. Our work assessed how the GGGGCC HRE elicits downregulation of FG Nups and identified whether FG Nup deficits contribute to the cellular defects observed in C9ORF72 ALS. Furthermore we attempted to rescue cellular impairment by reversing FG Nup deficits in C9ORF72 ALS cellular and Drosophila models. Through our understanding of nucleoporin deficits we may identify novel approaches to reversing cellular impairment in C9ORF72 ALS.

First Author: Diane Carlisle (Faculty)	Poster Session: pm
Presenting Author: Tanisha Singh (Postdoctoral)	Location: 3
Mentor/Lab: Diane Carlisle	Category: Neurology & Neurodegenerative Diseases
Department: Neurological Surgery	
Title: Characterizing Mitochondrial Dysfunction in Sporadic ALS Patient Motor Neurons	
Summary: In this project we investigate characterize dysfunction in human motor neurons from sporadic ALS patients.	
Abstract: Approximately 5-10% of ALS cases are familial (fALS). In fALS autosomal mostly dominant mutations have been reported in several genes such as C9orf72 (40%) SOD1 (20%) TAR DNA-binding protein-43 (TDP-43) (3%) FUS/TLS (5%) and TAF-15 (1%). Among the multiple proposed mechanisms based mainly on experimental in vivo and in vitro models a key role is attributed to the activation of mitochondrially mediated neuronal death signaling pathways. However the majority (90-95%) of ALS cases are sporadic (sALS). The pathobiology of sALS is largely unknown despite suspected genetic and environmental factors at play. In sALS patients no specific molecular biology characterization or timing of mitochondrial changes during neuronal maturation have been reported. Using established protocols we generated induced pluripotent stem cells (iPSCs) from sALS patients and differentiated iPSCs into neural progenitor and mature motor neurons (MNs). We examined mitochondrial parameters in sALS cells from all three developmental stages and compared them with controls. Our studies demonstrate that developmental stage plays a crucial role in the ALS phenotype in vitro and that these cells can be used to investigate mitochondrial dysfunction in sALS.	

First Author: Yunhong Huang (Postdoctoral)	Poster Session: pm
Presenting Author: Yunhong Huang (Postdoctoral)	Location: 4
Mentor/Lab: Amantha Thathiah	Category: Neurology & Neurodegenerative Diseases
Department: Department of Neurobiology	

Title: In vivo inactivation of β-arrestin 2 signaling in Alzheimer's disease

Summary: Alzheimer's disease (AD) is one of the most significant medical and societal challenges of our time and yet no current intervention strategies can halt or modify the underlying disease course. Our lab identified the orphan G protein-coupled receptor (GPCR) GPR3 as a primary modulator of AD pathology. The current study investigates the in vivo therapeutic modulation of GPR3 signaling to understand disease mechanisms and open a potentially novel avenue for therapeutic intervention in AD.

Abstract: Alzheimer's disease (AD) is one of the most significant medical and societal challenges of our time and yet no current intervention strategies can halt or modify the underlying disease course. Clinically AD is characterized by progressive memory loss personality disturbances and general cognitive decline. Neuropathologically AD is characterized by the accumulation of amyloid-β (Aβ) tau and neuroinflammation. Aβ is derived from proteolysis of the β-amyloid precursor protein (APP) following sequential cleavage by the β- and γ-secretases. G protein-coupled receptors (GPCRs) are involved in key neurotransmitter systems that are disrupted in AD patients and are also associated with multiple stages of APP proteolysis indicating an intimate association between GPCRs and the molecular pathways involved in AD. We identified the orphan GPCR GPR3 as a key modulator of γ-secretase activity and determined that β-arrestin 2 (βarr2) which belongs to a small family of multifunctional GPCR adaptor proteins specifically interacts with the γ-secretase complex and critically is required for the GPR3-mediated effect on Aβ generation. These results support the hypothesis that βarr2 is a critical link between GPCR dysfunction and Aβ generation in AD. Here we sought to determine the in vivo consequence of selective abrogation of βarr2-dependent signaling on amyloid pathology which is likely essential for triggering physiological and pathophysiological outcomes in mouse models of the disease. We utilized a CRISPR/Cas9-mediated genome editing strategy to introduce defined point mutations in the C-terminus of murine Gpr3 to interfere with the interaction between GPR3 and βarr2. These studies will provide the first demonstration of the in vivo consequence of selective modulation of βarr2-dependent signaling in AD pathogenesis. Results from these studies will not only address a major challenge in understanding disease mechanisms in AD they will also provide new avenues for the development of potential therapeutic targets to mitigate and/or halt the neurodegenerative changes observed in AD.

First Author: Matthew Phillips (Graduate)	Poster Session: pm
Presenting Author: Matthew Phillips (Graduate)	Location: 5
Mentor/Lab: Jon Johnson	Category: Neurology & Neurodegenerative Diseases
Department: Neuroscience	

Title: Characteristics of NMDA receptor channel block by the novel polycyclic amines RL-202 and RL-208

Summary: New drugs (the compounds RL-202 and RL-208) show similar biophysical properties to the Alzheimer's disease drug memantine sharing a conserved basic mechanism of action and overlapping binding sites in the NMDA receptor a brain protein necessary for learning and memory. Interestingly these drugs perform some forms of inhibition more strongly than memantine. Studying how RL-202 and RL-208 affect the NMDA receptor can help us further understand how channel blocking compounds function and will aid in future design of better drugs.

Abstract: NMDA receptors (NMDARs) are a class of ionotropic glutamate receptors (iGluRs) expressed at nearly all vertebrate synapses. NMDARs display a variety of properties unique amongst iGluRs including dependence upon co-agonists voltage-dependent Mg2+ block slow kinetics and permeability to Ca2+. NMDAR activity is critical for many types of synaptic plasticity and is a key player in memory formation and learning. Conversely aberrant NMDAR activation is implicated in a variety of nervous system disorders such as Alzheimer's disease and stroke. Pharmacological targeting of NMDARs with channel blockers has shown therapeutic promise for protection from excitotoxicity as well as the treatment of Alzheimer’s disease and major depressive disorder. Despite sharing similarities in binding site and mechanism of inhibition the clinical utility of NMDAR channel blockers with differing structure can vary dramatically. Further investigation into how channel blockers differentially affect receptor function may provide insight into their varying clinical efficacy and aid in future drug design. Here we provide pharmacological characterization and comparison of two novel NMDAR channel blockers the polycyclic amines RL-202 and RL-208 with the Alzheimer’s disease drug memantine (Mem). RL-202 and RL-208 were found to be voltage-dependent trapping channel blockers that possess similar IC50 values to Mem. Interestingly both RL-202 and RL-208 display stronger second site inhibition (SSI) a form of antagonism that involves drug binding to a site outside the NMDAR channel in the absence of agonist than Mem despite their similar structures and traditional potencies. Our findings suggest that these compounds could be useful tools for elucidating and differentiating mechanisms of NMDAR inhibition by channel blockers.

First Author: Nicholas Todd (Graduate)	Poster Session: pm
Presenting Author: Nicholas Todd (Graduate)	Location: 6
Mentor/Lab: Thathiah	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	

Title: G protein-coupled receptor kinases as a therapeutic target for Alzheimer's Disease

Summary: Of the top ten leading causes of death worldwide Alzheimer's disease (AD) is the only one that we cannot prevent cure or slow down. We identified the orphan G protein-coupled receptor (GPCR) GPR3 as a primary modulator of AD pathology. Here we investigate the pathophysiological role that GPCR kinases (GRKs) play in modulation of GPR3 function and disease progression. Results from these studies will not only address a major challenge in understanding disease mechanisms in AD they will also provide new avenues for the development of potential therapeutic targets to mitigate and/or halt the neurodegenerative changes observed in this devastating neurodegenerative disorder.

Abstract: Alzheimer's disease (AD) is characterized by the accumulation of aggregates of the amyloid-β (Aβ) peptide formed by sequential cleavage of the β-amyloid precursor protein (APP) by the β- and γ-secretases. Changes in APP and/or Aβ homeostasis lead to Aβ aggregation that critically contributes to the pathological abnormalities associated with AD. As such pharmacologically targeting of Aβ is one of the primary approaches investigated to treat AD. G protein-coupled receptors (GPCRs) are the most common target for therapeutic drug discovery. Several GPCRs have also been associated with multiple stages of APP proteolysis. Our lab identified the orphan GPCR GPR3 as a modulator of Aβ pathology. Furthermore we determined that the GPR3-mediated effect on Aβ generation requires the GPCR adaptor protein β-arrestin 2 (βarr2). GPCR kinases (GRKs) bind GPCRs upon ligand activation and phosphorylate GPCRs triggering βarr2 recruitment and subsequent downstream signaling. Significantly evidence suggests that levels of GRK2 and GRK5 are altered in the human AD brain. Despite these findings the putative involvement of GRKs in AD pathology has not been investigated in any context. Indeed identification of the GRKs involved in the modulation of GPR3 and βarr2 function could provide fundamental novel insight into the contribution of this important class of kinases under physiological and pathophysiological conditions. In preliminary studies genetic deletion of each of the four ubiquitously expressed GRKs namely GRKs 2 3 5 and 6 using a CRISPR/Cas9 genome-editing strategy indicates that GRKs 2 3 and 5 differentially regulate Aβ generation. We are currently testing the hypothesis that specific GRKs also regulate βarr2 recruitment to GPR3 and/or the phosphorylation status of GPR3 and the γ-secretase. Collectively these studies will determine the pathophysiological involvement of GRKs in the regulation of γ-secretase function establish which GRKs are involved in GPR3 phosphorylation and βarr2 recruitment and provide a potentially innovative therapeutic approach to treat AD.

First Author: Bistra Iordanova (Faculty)	Poster Session: pm
Presenting Author: Bistra Iordanova (Faculty)	Location: 7
Mentor/Lab: Vazquez	Category: Neurology & Neurodegenerative Diseases
Department: Bioengineering	

Title: In vivo NADH fluorescence imaging of double transgenic AD mice reveals chronic tissue hypoxia

Summary: Shedding light on the relationship between Alzheimer's disease (AD) oxygen metabolism and neurovascular deficits is the goal of this project. AD and vascular disease were traditionally considered separate conditions AD being caused by brain neurodegeneration and the vascular deficits caused by pathological changes in the blood vessels. Recently increasing evidence indicates that there is a connection between these two conditions. The relationship between AD and the neurovascular deficits is the focus of this project. The results can ultimately lead to new clinical therapies that target vascular and metabolic pathways to halt AD progression.

Abstract: Background: Vascular and metabolic dysfunctions are well known features of Alzheimer's Disease (AD) and they precede clinical dementia. Undoubtedly vascular changes are expected as amyloid accumulates in the arterial vessel walls in cerebral amyloid angiopathy (CAA) leading to the death of smooth muscle cells cerebral hypoperfusion and inadequate oxygen supply. These vascular events could also contribute to metabolic alterations in glucose homeostasis. High resolution in vivo study of the dynamic vascular and metabolic events may reveal which tissue regions and cell populations are affected and cast light on the mechanisms that contribute to AD pathogenesis. Methods: We used fluorescence imaging of nicotinamide adenine dinucleotide (NADH) as an intrinsic marker for cellular metabolic states and tissue oxygen supply in vivo. We resolved the tissue boundaries of NADH fluorescence in the cortex of transgenic AD mice (B6C3.Tg(APPswe-PSEN1de9) n=4 12-24 months old) and observed NADH pattern relative to vessels during hyperoxia and normoxia. We then used in vivo two-photon fluorescence microscopy together with cell-type specific labeling to determine the cellular origin of the intrinsic signal and the locality of CAA. Results: Reduction of oxygen supply from hyperoxia to normoxia produced no detectable changes in controls however AD mice showed characteristic NADH pattern (Figure 1A) indicative of reduced oxygen gradient and rise in glycolysis in tissues further away from the arterial oxygen supply. Areas around capillary beds showed decreased NADH signal. Two-photon imaging under the same conditions revealed numerous cells with increased signal (Figure 1B) and only some of those cells stained positive for the astrocyte marker Sulforhodamine-101 (Figure 1C). All AD mice had CAA and tissue plaques seen with Methoxy-X04 staining (Figure 1D) and there appeared to be no association of the NADH signal with the plaques location. Conclusion: In agreement with previous findings double transgenic AD mice display chronic tissue hypoxia. Our preliminary results also indicate that under those conditions a subset of cells may adapt by up-regulating glycolysis to overcome the deficient oxidative phosphorylation. The population of cells with increased NADH signal is likely a combination of neurons and glia. This work can lead to new strategies that target metabolic pathways to halt AD progression.

First Author: Yanjun Zhao (Postdoctoral)	Poster Session: pm
Presenting Author: Yanjun Zhao (Postdoctoral)	Location: 8
Mentor/Lab: Zak Wills	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	
Title: Amyloid Beta Peptides Block New Synapse Assembly by Nogo Receptor Mediated Inhibition of T-Type Calcium Channels	
Summary: Imaging and electrophysiological studies of Nogo receptor - Amyloid beta signaling in hippocampus	
Abstract: Compelling evidence links amyloid beta (Abeta) peptide accumulation in the brains of Alzheimer's disease (AD) patients with the emergence of learning and memory deficits; yet a clear understanding of the events that drive this synaptic pathology are lacking. We present evidence that neurons exposed to Abeta are unable to form new synapses resulting in learning deficits in vivo. We demonstrate the Nogo receptor family (NgR1-3) act as Abetareceptors mediating an inhibition of synapse assembly plasticity and learning. Live imaging studies reveal Abeta activates NgRs on the dendritic shaft of neurons triggering an inhibition of calcium signaling. We define T-type calcium channels as the target of Abata-NgR signaling mediating Abeta's inhibitory effects on calcium synapse assembly plasticity and learning. These studies highlight deficits in new synapse assembly as a potential initiator of cognitive pathology in AD and pinpoint calcium dysregulation mediated by NgRs and T-type channels as key components.	

First Author: Kevin Sullivan (Postdoctoral)	Poster Session: pm
Presenting Author: Kevin Sullivan (Postdoctoral)	Location: 9
Mentor/Lab: Mary Ganguli	Category: Neurology & Neurodegenerative Diseases
Department: Epidemiology	

Title: Dementia Incidence in Four Population-Based Cohorts: The MYHAT and MoVIES Studies

Summary: Several epidemiological studies worldwide have reported decreasing dementia incidence rates (new cases) for more recent birth cohorts (born after 1920) compared to earlier both cohorts (born before 1920). We aimed to examine dementia incidence rates in four birth cohorts (1902-1911 1912-1921 1922-1931 1932-1941) across two Western PA population-based epidemiological studies to see if we observed the same downwards trend.

Abstract: Several large epidemiological studies have reported a decrease in incident dementia rates for more recent birth cohorts particularly in developed nations. Examining dementia incidence trends have many implications for preventive interventions. Pooling data from two large prospective population-based epidemiological dementia studies in Western Pennsylvania between 1987-Present we were able to identify four birth cohorts: 1902-1911 (n=421) 1912-1921 (n=1399) 1922-1931 (n=1075) 1932-1941 (n=670). With a total starting sample of 3565 we examined the incident dementia rates (dementia defined as Clinical Dementia Rating ≥ 1) using a proportional hazards model based on birth cohort with adjustment for baseline age sex education and study. Cohort effects in dementia incidence rates were observed with the most recent cohort reference group (1932-1941) having significantly lower incidence dementia rates compared to all three of the earlier birth cohorts (p<.01). Additionally dementia incidence trended downwards from the earliest birth cohort (1902-1912) with each successive birth cohort. This trend was unexplained by adjustment for baseline age education sex or study. Data from our pooled population-based studies supports other reports of declining dementia incidence rates in more recent birth cohorts and that this decline is not due to differences in education sex baseline age or which of the two pooled studies the participant was in. Further investigations into risk factors that may account for this trend are necessary.

First Author: Fangzhou Cheng (Graduate)	Poster Session: pm
Presenting Author: Fangzhou Cheng (Graduate)	Location: 10
Mentor/Lab: Anne M. Robertson	Category: Neurology & Neurodegenerative Diseases
Department: Mechanical Engineering	

Title: Understand the structural mechanism of cerebral aneurysm bleb: ruptured vs. stable. Report of two cases.

Summary: The rupture risk of cerebral aneurysm is strongly correlated to the appearance of aneurysm bleb. The purpose of this study is to gain insight into the structural mechanism of stable and ruptured blebs.

Abstract: Aneurysm blebs are outward surface protrusions that form on the side of aneurysm walls. They are speculated to be bulging weakened areas that reduce the tensile stress in the aneurysm wall. Even though this hypothesis suggests a protective role against aneurysm rupture a strong correlation has been found between the aneurysm blebs and rupture. However despite this established association little is known about the remodeling mechanisms within the aneurysm bleb. To gain insight into these mechanisms we analyzed blebs in two aneurysms - one stable and one ruptured. Multiphoton microscopy (MPM) was used to obtain the collagen fiber structure of the blebs and their parent aneurysm wall. Collagen fiber recruitment and orientation distribution were directly measured from the MPM images. The collagen fiber orientation distribution was mapped back to the 3D geometry obtained by micro-CT and compared with the stress distribution calculated using a customized finite element code. The relationship between wall architecture and intramural stresses were compared in the ruptured and unruptured aneurysm blebs and different structural mechanisms explained.

First Author: Michael Durka (Graduate)	Poster Session: pm
Presenting Author: Michael Durka (Graduate)	Location: 11
Mentor/Lab: Anne M. Robertson	Category: Neurology & Neurodegenerative Diseases
Department: Department of Mechanical Engineering and Materials Science	

Title: Oxygen Transport in Cerebral Aneurysms

Summary: Cerebral aneurysms can be lethal or severely debilitating if they rupture but what exactly causes them to weaken to the point of rupturing is not well understood. This study utilized computer simulation techniques to analyze the transport of oxygen from the blood to the interior of the aneurysm wall - something which cannot be done clinically due to the limits of current technology. The goal of this study was to determine whether the unusual blood flow patterns in a cerebral aneurysm (relative to the normal blood flow patterns in a normal artery) diminish the amount of oxygen transported to the aneurysm wall to a point which the lack of oxygen could potentially cause damage to the wall tissues.

Abstract: Cerebral aneurysms are abnormal balloon-like structures in brain arteries which are often mechanically inferior to a healthy artery in that their yield strength is significantly reduced. This reduction in yield strength can lead to rupture which often has debilitating if not lethal consequences. Surgical intervention though a potential solution at preventing such an event carries its own risks to a patient which sometimes exceed the natural risk of rupture. It's therefore critical to be able to reliably determine the propensity for rupture; unfortunately this is not yet possible with current minimal-risk non-invasive techniques. Furthermore the exact cause(s) of this condition is not fully understood. While the impacts of fluid-influenced mechanical factors such as wall shear stress (WSS) magnitude (low high or both) and direction (temporally stable or unstable) as well as intra-aneurysmal blood flow structure on wall degradation have been heavily studied little work has been done (with cerebral aneurysms) to study the influence of fluid-influenced chemical-based factors such as mass transport of oxygen; particularly the impact of the abnormal intra-aneurysmal flow pattern (relative to a healthy artery) on the effectiveness of nourishment (or lack thereof) to the aneurysm wall. Hypoxia has already been implicated in the development of abdominal aortic aneurysms; therefore it is reasonable to explore the same effect in the context of cerebral aneurysms. We therefore conducted a computational study of oxygen transport in two cerebral aneurysms having identical parent vessels but different aneurysm geometries. The impact of geometry on flow structure and mass transport was then analyzed. Qualitative relationships between oxygen transport and WSS were also explored. The study then yielded an assessment as to the degree to which aneurysm geometry can influence the concentration of molecular oxygen within the aneurysm wall. Such information in larger future studies could aid in further understanding the disease

First Author: Chao Sang (Graduate)	Poster Session: pm
Presenting Author: Chao Sang (Graduate)	Location: 12
Mentor/Lab: Anne Robertson	Category: Neurology & Neurodegenerative Diseases
Department: Mechanical Engineering and Materials Science	

Title: MECHANICAL RESPONSE AND FIBER REMODELING IN ELASTASE-INDUCED RABBIT ANEURYSMS

Summary: As in an evolving human cerebral aneurysm the rabbit aneurysm wall experiences changing tensile loads after creation and must adapt its extracellular matrix. The average wall strength increased over time suggesting effective fiber remodeling in adaptation to the increased axial load. The medial layer demonstrated a transition from largely circumferential loading to multiple fiber directions better suited to manage the biaxial loading found in the aneurysm wall.

Abstract: An intracranial aneurysm (IA) is most commonly a saccular enlargement in the wall of a cerebral artery. Aneurysm rupture is associated with high morbidity and mortality and hence there is a pressing need to better understand disease progression and to identify clinically useful metrics for assessment of rupture risk. It is commonly accepted that stress factors such as abnormal hemodynamics can lead to wall degradation that sometimes present in the clinic as changes to the aneurysm shape and size. However in most cases such longitudinal information is not available and aneurysm size is used for risk assessment. Human intracranial aneurysm samples can be obtained following treatment by surgical clipping and have provided valuable information about the heterogeneity in the aneurysm wall among patients. Recent studies have addressed the relationship between hemodynamics and changes to the aneurysm wall. A challenge is that harvested aneurysm tissue from patients only represents one time point in the pathology. Animal models for IAs provide a means of studying the evolving aneurysm wall. In this work we used an elastase induce aneurysm model in rabbits to study progressive changes in wall structure and mechanical properties. As in an evolving human cerebral aneurysm the rabbit aneurysm wall experiences changing tensile loads after creation and must adapt its extracellular matrix. We used multi-photon microscopy to measure collagen fiber remodeling and uniaxial testing to evaluate the corresponding changes to mechanical properties. The average wall strength increased over time suggesting effective fiber remodeling in adaptation to the increased axial load. This remodeling occurred in a non-homogeneous manner across the wall thickness. The medial layer demonstrated a transition from largely circumferential loading to multiple fiber directions better suited to manage the biaxial loading found in the aneurysm wall. In the future the rabbit model can be used to evaluate cellular activities responsible for these changes and to test pharmacological treatments that augment these changes.

First Author: Anne Robertson (Faculty)	Poster Session: pm
Presenting Author: Anne Robertson (Faculty)	Location: 13
Mentor/Lab: Robertson	Category: Neurology & Neurodegenerative Diseases
Department: Mechanical Engineering and Materials Science	

Title: ROLE OF CALCIFICATION IN ANEURYSM FAILURE- A CASE STUDY

Summary: The rupture of a brain aneurysm has high mortality and disability rates. To date the cause of rupture is poorly understood limiting possibilities for screening patients for aneurysms at risk for rupture. The objective of this work is to use use mutiple imaging and mechnical testing modalities to assess the role of calcification in aneurysm rupture.

Abstract: Intracranial aneurysms are believed to exist in approximately 5% of the adult population. While rupture is relatively rare intracranial hemorrhage due to rupture has devastating effects with high mortality and disability rates. Since risks associated with aneurysm treatment can exceed the natural risk of rupture there is an urgent need for a reliable method to identify fragile aneurysms at risk of rupture from those that can be safely monitored. In order to better understand rupture risk it is valuable to consider the wall prior to rupture since substantial biological geometric and structural changes can occur after rupture and possibly even days or even weeks prior to rupture. In our earlier work we introduced a classification system for dividing unruptured cerebral aneurysm tissue into robust and vulnerable groups. Here we build on this work and introduce an approach for exploring wall vulnerability using micro-CT imaging mechanical testing and computational studies. We consider a case study of an unruptured cerebral aneurysm and explore the source of wall vulnerability.

First Author: Ronald Fortunato (Graduate)	Poster Session: pm
Presenting Author: Ronald Fortunato (Graduate)	Location: 14
Mentor/Lab: Spandan Maiti	Category: Neurology & Neurodegenerative Diseases
Department: Department of Mechanical Engineering and Material Science and Bioengineering	

Title: COMPUTATIONAL STUDY OF UNIAXIAL TENSION TESTING OF SMALL SOFT TISSUE SPECIMEN

Summary: In this article we investigate the material properties used in uniaxial tensile grips that will ensure development of uniaxial stress state within a tissue sample and failure of the tissue in the region where uniaxial conditions prevail based on a finite element model. We also model failure and parametrically vary tissue strength and toughness to quantify failure mode of the tissue. Property-function relationship of the wall tissue will enhance our understanding about different clinical scenarios where some aneurysms fail catastrophically while others gradually progress towards rupture.

Abstract: Uniaxial testing is the most popular method for the evaluation of biomechanical properties of soft tissue. In this method a tissue specimen is fixed between two grips and stretched with a known displacement in one direction while the load borne by the specimen is recorded. The load-displacement data provides the constitutive behavior of the tissue. Often the specimen is also stretched until failure to ascertain the uniaxial strength of the tissue. For accurate evaluation of the material properties however uniform stress transmission within the tissue needs to be attained. The fixity at the tissue-grip interface is known to give rise to localized stress concentrations or even tissue damage that may provide erroneous uniaxial data. The standard practice to alleviate this problem is to attach the tissue to pieces of an intervening material typically sandpaper or cardboard glued to the metallic grips. However no analysis exists in the literature to ascertain whether this arrangement results in uniform stress distribution in the vicinity of the grips. For this study we present a detailed computational study of the effect of grip design and tissue shape on the stress state of the soft tissue specimen. Concurrently we studied the effect of tissue strength and toughness on failure. We developed an image derived finite element model of a dog bone shaped tissue specimen attached to steel grips through a thin layer of soft material. The grips were first clamped down on the specimen with a specified pressure and then uniaxial displacement was applied to one of the clamps. The strength and toughness was parametrically varied to observe the evolution of tissue damage that would lead to tissue failure. We computationally found that insertion of a soft rubber layer between the steel grips and cerebral artery tissue specimen resulted in uniform uniaxial stress near the midlength of the specimen while no stress concentration was observed near the grips. In addition damage was also localized in the midregion of the specimen. These results are expected to provide guidelines for proper design of grips for the uniaxial testing apparatus for testing of soft tissues in general and cerebral arterial wall tissue in particular.

First Author: C. Elizabeth Shaaban (Graduate)	Poster Session: pm
Presenting Author: C. Elizabeth Shaaban (Graduate)	Location: 15
Mentor/Lab: Dr. Caterina Rosano	Category: Neurology & Neurodegenerative Diseases
Department: Epidemiology	

Title: Response of venous-side microvasculature in older adults to physical activity intervention: A study at 7T

Summary: We found that a structured walking routine and increases in brain-derived neurotrophic factor a growth factor can beneficially impact the brain's small veins even among very old adults. These may be promising ways to prevent or treat Alzheimer's disease or small vessel disease in the brain but future studies will be needed to confirm this.

Abstract: BACKROUND: We have recently shown that tortuosity of brain small veins are cross-sectionally associated with having at least one APOE4 allele and with lower levels of vascular endothelial growth factor (VEGF) (Shaaban et al. 2017). Prior work has shown that PA can lower severity of brain small vessel disease (SVD) and risk of Alzheimer's disease (AD). Here we test the hypothesis that physical activity reduces tortuosity of brain small veins. We also explore the effects of VEGF and brain-derived neurotrophic factor (BDNF) because of their beneficial effects on the vasculature. METHODS: Participants of the LIFE study (N=14 7 in each arm) mean age 77 (range 70-86) 85.7% female 42.9% non-white) were randomly assigned to a 24-month program of center-based walking 2x/week (PA) or a health education (HE) program. Moderate levels of PA were objectively measured by accelerometry as cumulative minutes/day spent at baseline and at 6 12 and 24 months. APOE4 genotype was determined via TaqMan and Pyrosequencing. Vein length and serum levels of vascular endothelial growth factor (VEGF) and brain-derived neurotrophic factor (BDNF) were measured at baseline and after 24 months. Lengths of tortuous and straight veins within a periventricular region of interest were measured and tortuosity ratio of total tortuous length to total straight length was considered a marker of SVD. Percent change from baseline to 24 months was computed for all measures of interest. Spearman correlations assessed relationships of percent change in venular markers with PA and molecular markers with alpha of 0.10. RESULTS: Intervention groups did not differ significantly at baseline ($p>0.05$). Greater cumulative PA by accelerometry predicted decrease in tortuosity ratio independent of arm assignment (rho= -0.533 $p=0.06$). Increase in BDNF also predicted decrease in tortuosity ratio independent of arm assignment (rho= -0.614 $p=0.03$). Neither change in VEGF nor APOE4 allele were related to change over time in tortuosity ratio ($p>0.10$). CONCLUSIONS: Greater cumulative PA and increased BDNF levels were associated with reduced tortuosity ratio. A structured walking intervention and greater BDNF levels may beneficially impact the venous-side microvasculature even among very old adults. Future studies should clarify the long-term effects of PA and BDNF on SVD and progression to AD.

First Author: Harman Ghuman (Graduate)	Poster Session: pm
Presenting Author: Harman Ghuman (Graduate)	Location: 16
Mentor/Lab: Dr. Mike Modo	Category: Neurology & Neurodegenerative Diseases
Department: Bioengineering	

Title: ECM hydrogel injection for the treatment of stroke

Summary: Functional replacement of the damaged brain tissue after stroke remains a major therapeutic challenge. Here we demonstrate a long term retention of ECM hydrogel in the stroke cavity that promotes influx of host cells into the biomaterial and eventually leading to a reduction in lesion volume over 3 months.

Abstract: Stroke is the leading cause of adult disability and a significant effort is underway to develop therapies to repair the damaged tissue. One of the key challenges in treating chronic stroke is the dramatic loss of brain tissue and the formation of a cavity filled with extracellular fluid (ECF) and cell debris. Extracellular matrix (ECM) constitutes 20% of brain tissue volume. Biomaterials composed of mammalian ECM promote constructive tissue remodeling with minimal scar formation in peripheral tissue and organs. However the biodegradation and functional effect of injecting a large volume of ECM hydrogel into the brain are unknown. The current study therefore aimed to determine if biodegradation occurs and if ECM remodeling will affect the behavioral deficits of animals with stroke damage. At an 8 mg/mL concentration ECM hydrogel has rheological properties similar to brain tissue. It can be formulated in a fluid phase at room temperature while forming hydrogels at body temperature. Two weeks post-stroke Magnetic Resonance Imaging-defined lesion volume equivalents of ECM was injected into the lesion cavity of stroke rats. A battery of behavioral tests including Grip Strength Bilateral Asymmetry Test (BAT) Footfault and Rotameter were performed at pre-treatment 1 4 and 12 weeks post-treatment for control (n=14) untreated (n=11) and ECM-treated (n=11) groups. Retention gelation and biodegradation of the ECM as well as host cell invasion and phenotype were analyzed at 12 weeks post-injection using immunohistochemistry. Brain tissue deformation analysis using T2-weighted MRI scans indicated a 10% decrease in whole brain tissue volume 2-fold increase in ventricle size a 10% midline shift and 30% decrease in tissue in the stroke-affected hemispheres over 12 weeks. There was no significant difference between untreated and treated groups. Behavioral tests indicated a functional impairment that was not affected by the injection of a large volume of ECM into the cavity. ECM showed a robust gelation and retention in the lesion cavity with a 30% decrease in volume over 12 weeks. A significant host cell invasion into the ECM hydrogel was seen with an average of 72000 cells present within the hydrogel. Monocytes accounted for 55% of the total invading cells and expressed a neutral M1/M2 (CD86/206) phenotype indicating a shift from the acute inflammatory phase to an ECM remodeling phase. Significant proportions of oligodendrocyte progenitor cells (30%) and endothelial cells (4-5%) essential for repopulation of the neural tissue were also present. This characterization demonstrates that an ECM hydrogel can be readily injected and retained within the lesion cavity while promoting an acute endogenous repair response without deleterious effects. A time course study with varying ECM concentrations is necessary to determine the optimal rate of in vivo biodegradation to further improve the endogenous repair processes.

First Author: Chenxiao Tang (Graduate)	Poster Session: pm
Presenting Author: Chenxiao Tang (Graduate)	Location: 17
Mentor/Lab: Samuel Poloyac	Category: Neurology & Neurodegenerative Diseases
Department: Pharmaceutical Sciences	

Title: Screening 20-HETE Formation Inhibitors in Microsomal Incubates Using UPLC-MS/MS

Summary: 20-HETE formation inhibitors were screened in in vitro system. Lead compounds with better solubility metabolic stability and potency could be used in preclinical animal model to evaluate its PK/PD.

Abstract: Introduction: 20-hydroxyeicosatetraenoic acid (20-HETE) is a metabolite of arachidonic acid (AA) by CYP4A11 and CYP4F2 in human with potent microvascular constriction activity. Inhibition of 20-HETE formation has neuroprotective effect in subarachnoid hemorrhage (SAH) cardiac arrest and thromboembolic stroke preclinical models. Clinical evidence shows that high level of 20-HETE is associated with three-fold increased mortality and unfavorable outcomes in SAH patients. These findings suggest that inhibition of 20-HETE formation is a potential therapeutic strategy for neuroprotection after brain injury. HET0016 a commonly used 20-HETE inhibitor is not suitable for clinical use due to its poor solubility and short half-life. At this point a clinically relevant 20-HETE inhibitor is not available to be evaluated as a therapeutic intervention. Hypothesis: Drug-like compounds that inhibit 20-HETE formation can produce neuroprotective effect in secondary brain injury by improving CBF and attenuating ischemic brain damage. Methods: Test compounds were obtained either via virtual screening against a CYP4F2 homology model or from scaffold hopping from structures of known inhibitors. Four different types of microsomal systems including human liver microsome (HLM) recombinant CYP4F2 (rCYP4F2) rat liver microsome (RLM) and rat kidney microsome (RKM) were used for microsomal incubations. AA was incubated in microsomes with/without compound for 20 min. 20-HETE formation rate was quantified using a validated UPLC-MS/MS assay and normalized by vehicle control group. Other eicosanoids including 15- 12-HETEs epoxyeicosatrienoic acids (EETs) and dihydroxyeicosatrienoic acids (DiHETs) were monitored simultaneously. Selected compounds metabolic stability was tested in HLM throughout 60-min incubation time the remaining compound was measured by UPLC-MS/MS and normalized to corresponding 0-min values. Results: We identified UPMP 10 as the hit compound. The IC50 of UPMP10 in HLM is 443.4nM. UPMP19 showed improved 20-HETE inhibitory effect with an IC50 of 187.1nM. Both UPMP10 and UPMP19 did not inhibit EETs or DiHETs formation up to 50000 and 10000 nM respectively. UPMP10 and 19 were more stable with 91.4±11.0% and 100.4±1.7% remaining compound at 30min in HLM compared to 35.1±5.7% of TS_24. After structure modification UPMP22 is the most potent compound with a IC50 of 49.56 nM. None of the six compounds inhibit epoxygenase pathway of AA. All the compounds were slowly metabolized in HLM throughout 60 min incubation time. UPMP22 has 85.4±1.51% remaining compound at 30 min time point. Conclusion: These results suggested that UPMP22 is a potent selective metabolically stable 20-HETE formation inhibitor. It can serve as preclinical lead for further structure modifications that may lead to novel 20-HETE formation inhibitors.

First Author: Dana Jorgensen (Graduate)	Poster Session: pm
Presenting Author: Dana Jorgensen (Graduate)	Location: 18
Mentor/Lab: Gianaros / Rosano	Category: Neurology & Neurodegenerative Diseases
Department: Epidemiology	

Title: Racial Differences in Brain Health at Midlife and the Potential Mediating Role of Cardiometabolic Risk.

Summary: Blacks are at a higher risk of stroke and developing dementia than whites but it remains to be determined whether differences in brain health are evident at midlife. Here we found several racial differences in brain health and that cardiometabolic risk was a partial mediator for the relationship between race and cortical surface area. These results have implications for understanding the pathways by which race may impact brain health prior to the onset of stroke and other clinical outcomes later in life.

Abstract: Introduction Blacks are at a higher risk of stroke and developing dementia than whites [4 5]. However much of what is known about racial differences in brain health is exclusive to those >65 years old and it remains to be determined whether relationships between race and brain health are apparent in midlife. Here we examined racial differences in brain health at midlife and tested whether cardiometabolic risk (CMR) statistically mediated any observed differences. Methods 747 community volunteers (20.6% black) aged 30–54 years old underwent neuroimaging to assess brain morphology and cerebral blood flow (CBF). Components of a composite CMR score included: body mass index waist circumference high-density lipoproteins triglycerides glucose insulin SBP and DBP. Results After adjustment for demographics and socioeconomic status blacks exhibited a significantly smaller hippocampus less cortical surface area and a thinner cerebral cortex than whites. We observed no significant differences in CBF. Mediation models showed that CMR partially mediated the association of race with cortical surface area. Conclusions Race differences in brain health are evident in midlife. CMR partially mediated the relationship between race and cortical surface area. These results have implications for understanding the pathways by which race may impact brain health prior to the onset of stroke and other clinical outcomes later in life.

First Author: Yalikun Suofu (Postdoctoral)	Poster Session: pm
Presenting Author: Yalikun Suofu (Postdoctoral)	Location: 19
Mentor/Lab: Friedlander	Category: Neurology & Neurodegenerative Diseases
Department: Neurological Surgery	

Title: Dual role of mitochondria in producing melatonin and driving GPCR signaling to block cytochrome c release

Summary: Melatonin is exclusively produced in mitochondria. Mitochondria membrane melatonin receptor type 1 respond to melatonin by activating heterotrimeric G proteins located in the mitochondria intermembrane space and inhibit stress-mediated cytochrome c release. Therefore the signaling pathway contributes to neuroprotection from ischemia-induced brain injury.

Abstract: G protein-coupled receptors (GPCRs) are classically characterized as cell-surface receptors transmitting extracellular signals into cells. Here we show that central components of a GPCR signaling system comprised of the melatonin type 1 receptor (MT1) its associated G protein and β-arrestins are on and within neuronal mitochondria. We discovered that the ligand melatonin is exclusively synthesized in the mitochondrial matrix and released by the organelle activating the mitochondrial MT1 signal-transduction pathway inhibiting stress-mediated cytochrome c release and caspase activation. These findings coupled with our observation that mitochondrial MT1 overexpression reduces ischemic brain injury in mice delineate a mitochondrial GPCR mechanism contributing to the neuroprotective action of melatonin. We propose a new term "automitocrine" analogous to "autocrine" when a similar phenomenon occurs at the cellular level to describe this unexpected intracellular organelle ligand-receptor pathway that opens a new research avenue investigating mitochondrial GPCR biology.

First Author: Victor Van Laar (Faculty)	Poster Session: pm
Presenting Author: Victor Van Laar (Faculty)	Location: 20
Mentor/Lab: Berman Lab	Category: Neurology & Neurodegenerative Diseases
Department: Neurology	

Title: Mitochondrial Mitofilin as a Novel Therapeutic Target for Parkinson's Disease

Summary: Currently there are no therapies for Parkinson's disease patients that alter or halt disease progression. Mitofilin a protein crucial for regulating mitochondrial function is an intriguing target for researching neuroprotective therapies and we have evidence that mitofilin overexpression is protective against Parkinson's-related neurotoxins in vitro. In this study we provide the first characterization of mitofilin in Parkinson's brain and begin evaluating mitofilin as a potential target for researching neuroprotective therapeutic treatments for Parkinson's disease.

Abstract: Parkinson's disease (PD) is the most common neurodegenerative movement disorder affecting 1% of people over the age of 65. At present there is no cure for PD and available treatments only address disease symptoms. Research is needed to identify novel targets for the development of neuroprotective therapies that will hinder or halt the progressive neuron loss in PD. Mitochondria are a major focus for PD research as mitochondrial dysfunction is a known contributor to PD pathophysiology. We have identified mitochondrial mitofilin a protein which functions as a unique nexus for regulating mitochondrial function and cellular stress response as a promising target for study. Mitofilin also known as mic60 is a scaffolding protein of the inner mitochondrial membrane and is critical for maintaining mitochondrial membrane structure and function. Mitofilin/mic60 also interacts with and regulates PINK1 a mitochondrial protein integral in signaling damaged mitochondria for degradation and associated with a heritable form of PD. Loss of mitofilin/mic60 has severely detrimental effects on mitochondrial morphology and respiration. Mitofilin/mic60 is also highly susceptible to oxidative stress. We and others have shown that mitofilin/mic60 protein levels are decreased in dopaminergic cells in models of PD. We have also previously shown that mitofilin/mic60 is a target for modification by oxidized dopamine the neurotransmitter used by the substantia nigral neurons that are uniquely vulnerable to PD. Further we found that a specific loss of mitofilin/mic60 in dopaminergic cells in vitro exacerbated cellular vulnerability and impaired respiratory capacity in response to rotenone a pesticide and mitochondrial Complex I inhibitor associated with increased PD risk. Conversely overexpression of mitofilin/mic60 promoted cellular survival and mitochondrial respiration. These results suggest that altering levels of mitofilin/mic60 in dopaminergic neuronal cells significantly affects both mitochondrial homeostasis and cellular vulnerability to PD-relevant stressors. We are now investigating mitofilin/mic60 for its role in PD pathogenesis and its neuroprotective potential. We have carried out an initial analysis of mitofilin/mic60 expression in PD patient brain. Post-mortem tissue from PD patient and age-matched control brains were immunohistochemically analyzed for mitofilin/mic60 expression level and cellular localization using confocal microscopy. To our knowledge this is the first such characterization of mitofilin/mic60 in human PD brain. We have also developed an adeno-associated viral vector for overexpression of mitofilin/mic60 in dopamine neurons in vivo which we will use to begin examining the neuroprotective properties of mitofilin/mic60. Our ultimate goal is to assess the neuroprotective capabilities of mitofilin/mic60 in vivo in preclinical models of PD.

First Author: Wenting Xie (Graduate)	Poster Session: pm
Presenting Author: Wenting Xie (Graduate)	Location: 21
Mentor/Lab: Edward A. Burton	Category: Neurology & Neurodegenerative Diseases
Department: PIND	

Title: Mitochondrial-Telomere ROS Cross-Talk in Parkinson's Disease

Summary: We hypothesize that ROS cross-talk induced a self-perpetuating cycles of damage between telomeres and mitochondria that underlies neurodegeneration in PD.To test this we generated transgenic zebrafish models in which we can uncouple telomeric and mitochondrial damage in the relevant disease-susceptible dopaminergic neurons in vivo using a novel chemoptogenetic ablation method.

Abstract: Mitochondrial reactive oxygen species (ROS) are regarded central to Parkinson's disease (PD) pathogenesis; however the role of mitochondrial oxidative damage to telomeres is unknown. Recent evidence suggests that telomeric dysfunction can result in mitochondrial defects. We hypothesize that ROS cross-talk induced a self-perpetuating cycles of damage between telomeres and mitochondria that underlies neurodegeneration in PD. To test this we generated transgenic zebrafish models in which we can uncouple telomeric and mitochondrial damage in the relevant disease-susceptible dopaminergic neurons in vivo using a novel chemoptogenetic ablation method. The method allows regulated generation of singlet oxygen in specific cellular locations. Since the effective range of the short-lived singlet oxygen is extremely small this results in oxidative damage to surrounding cellular components with a remarkable organelle-level degree of spatial resolution and with graded severity dictated by light dose. This new technology will enable us to test our hypothesis by inducing selective damage at mitochondria or telomeres while measuring ROS flux and dysfunction at both sites. Our initial data provide proof of concept that we can induce both functional and morphological changes both acutely and chronically in mitochondria targeted by our novel chemoptogenetic approach in zebrafish neurons in vivo resulting in neurological phenotypes.

First Author: De Miranda Briana (Postdoctoral)	Poster Session: pm
Presenting Author: Briana De Miranda (Postdoctoral)	Location: 22
Mentor/Lab: Greenamyre	Category: Neurology & Neurodegenerative Diseases
Department: Neurology	

Title: Sex differences in sensitivity to rotenone reflect male-to-female ratios in human Parkinson's disease incidence

Summary: Parkinson's disease affects males approximately 1.5 times more frequently than females however the reason for this is unknown. Animal models of PD rarely take into consideration sex as a variable therefore we examined the differences between male and female Lewis rats in the rotenone model of PD. Similar to human data females were resistant to rotenone degeneration and required a higher dose to produce equivalent pathology observed in male rats.

Abstract: The male to female odds ratio for incidence of Parkinson's disease (PD) is 1.49 indicating that sex differences likely play a role in the pathogenesis of the disease. Animal modeling of PD however rarely uses sex as a variable when examining neurodegeneration possibly overlooking important etiological factors. Rotenone an organic pesticide and prototypical mitochondrial complex I inhibitor reliably reproduces parkinsonism in rats including motor behavioral deficits of postural instability rigidity and bradykinesia. In addition rotenone causes the selective neurodegeneration of dopamine neurons in the substantia nigra (SN) and their terminal projections in the striatum (ST) endogenous alpha-synuclein accumulation microglial activation and changes in iron metabolism. To date the rotenone model has primarily been utilized in adult male Lewis rats however our pilot studies in adult female Lewis rats using the same dose of rotenone (2.8 mg/kg i.p.) did not yield equivalent motor behavioral changes nor dopamine neuron loss or brain pathology. Therefore we postulated that female rats may be less vulnerable to rotenone-induced neurodegeneration and would require a higher dose of rotenone to induce the parkinsonian morbidities observed in male rats. To this end we generated a dose-response using 2.8 mg/kg 3.2 mg/kg or 3.6 mg/kg (daily i.p.) of rotenone in female Lewis rats with one additional group receiving BID dosing (1.6 mg/kg total 3.2 mg/kg) of rotenone. Female rats receiving 3.2 mg/kg 1.6 mg/kg (BID) and 3.6 mg/kg rotenone had a significant loss of dopamine neurons within the SN as assessed by stereology accompanied by a loss of tyrosine hydroxylase-positive terminals in the ST. Significant microglial activation within the SN was observed in only the 1.6 mg/kg BID and 3.6 mg.kg group compared to a marked activation of microglia in male rats given 2.8 mg/kg. The transferrin receptor (TfR1) was measured as an indicator of cell surface iron binding and was significantly increased in male rats receiving 2.8 mg/kg of rotenone but did not result in a significant increase in female rats across any dose. Ferritin an iron binding protein expressed predominately in oligodendrocytes within the SN was significantly preserved in female rats following rotenone exposure (all doses) indicating that females may have better iron storage capacity following neurotoxic insult. Taken together these data indicate that female rats require a higher dose of rotenone to produce equivalent neurodegeneration in the rotenone PD model an effect that parallels human data of a higher prevalence of PD in males and highlights the importance of using female animals when experimentally modeling PD pathogenesis.

First Author: Amber Van Laar (Faculty)	Poster Session: pm
Presenting Author: Amber Van Laar (Faculty)	Location: 23
Mentor/Lab: Greenamyre Lab	Category: Neurology & Neurodegenerative Diseases
Department: Neurology	

Title: Progressive parkinsonism in rats following brief rotenone exposure: a novel model of Parkinson's disease

Summary: A more predictive and accurate model of Parkinson's disease is needed to facilitate the development of disease-modifying therapies. In this study we have developed and characterized a novel progressive animal model of Parkinson's disease. A key distinction of this model is the ability to test new possible therapies once the disease process is underway or even after symptom onset which is directly relevant to the Parkinson's disease patients in the clinic.

Abstract: A major barrier in treatment advancement for Parkinson's disease (PD) has been the lack of preclinical models that recapitulate the complexities of human PD with fidelity. The need for parkinsonian models with greater clinical predictive value has never been greater. Rotenone – a pesticide linked to increased PD risk and a potent inhibitor of mitochondrial respiration – has been a useful tool in PD research. Rotenone exposure has previously been demonstrated to produce a parkinsonian behavioral phenotype in rats associated with nigrostriatal degeneration when administered chronically. We have now found that just a brief exposure to rotenone triggers a downstream cascade of neurodegenerative events with progressive development of behavioral and neuropathological features analogous to human PD. Wildtype aged Lewis rats (6-9mo) were administered rotenone (i.p.) for only 5 days. The rats developed a parkinsonian phenotype during rotenone treatment but within 2 weeks after discontinuation of rotenone all rats recovered to their behavioral baseline where they remained until about 10 weeks. After this period of neurologic normalcy all rats spontaneously developed mild parkinsonian behavioral features that slowly progressed over the next 3-4 months. Immunohistochemical analyses revealed that during the behaviorally quiescent period while animals are at baseline nigral dopaminergic neurons began to accumulate alpha-synuclein which gradually begins to consolidate into Lewy body-like inclusions by 3 months when the parkinsonian phenotype returns. Microglial activation accompanies the accumulation of alpha-synuclein and loss of nigral dopamine neurons which indolently progresses over several months. Alpha-synuclein accumulation was also found outside of the nigrostriatal system including cortex and hippocampus in rats aged out to 9 months after the start of rotenone. We propose that this delayed rotenone model with a progressive endogenous alpha-synucleinopathy provides a more clinically predictive parkinsonian model to rigorously investigate PD-relevant disease mechanisms and potential therapeutics. A key advantage to this model is the delay of parkinsonian symptom onset after the brief rotenone exposure providing an opportunity to evaluate neuropathogenic mechanisms and therapeutic strategies both before and after symptom onset. The spontaneous development of symptoms that progresses over months - akin to human PD - allows testing of therapeutic interventions at multiple clinically-relevant time points. The prolonged survival after symptom development also allows for evaluation of therapeutic response over a period of months. The delayed parkinsonian rotenone model stands to serve as a preclinical and neurobiological surrogate for human PD. This new PD model provides for more accurate and efficient assessment of potential therapeutics thereby promoting the translation of impactful treatments more readily into clinical practice.

First Author: Joe Brague (Postdoctoral)	Poster Session: pm
Presenting Author: Joe Brague (Postdoctoral)	Location: 24
Mentor/Lab: Rebecca Seal	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	

Title: VGLUT3 Knockout Mice Show Increased Dopamine Synthesis Spine Density and Normal Motor Behavior in a Parkinson's Disease Model

Summary: Parkinson's Disease (PD) is characterized by a loss of dopamine which impacts structural and functional changes of neurons and ultimately leads to the debilitating loss of motor functions. Recently our lab reported that a genetically modified mouse lacking a specific excitatory transporter showed an increase in dopamine and the number of neuronal connections in PD neuronal circuitry and interestingly showed normal motor behavior in the PD model. This poster highlights these exciting findings and outlines an experimental plan to rescue the motor deficits seen in parkinsonian mice and ultimately in humans.

Abstract: Joe C. Brague Christopher B. Divito Rebecca P. Seal* Department of Neurobiology University of Pittsburgh Parkinson's Disease (PD) is a progressive and debilitating disorder of the nervous system afflicting approximately ten million people worldwide. Symptoms including postural instability and slowed gait stemming from the death of dopamine (DA) neurons of the substantia nigra pars compacta (SNpC). These DA neurons densely innervate the dorsal striatum and profoundly influence motor function through actions of medium spiny neurons (MSN) the major projection neurons of the striatum. Loss of striatal DA in PD is thought to affect motor behavior by altering direct (go) and indirect (no-go) pathway output of MSNs. Understanding more precisely how loss of DA affects basal ganglia motor circuits will greatly expand treatment options for patients suffering from PD. In this study we explore the role of the vesicular glutamate transporter 3 (VGLUT3) one of three proteins responsible for the uptake of glutamate into synaptic vesicles in DA signaling and motor behavior in healthy and Parkinsonian animals. We recently showed that mice lacking VGLUT3 (Vglut3-/-) have increased DA synthesis and release in the striatum during the waking cycle. Additionally the density of immature spines on direct pathway MSNs is also increased during this time. When tested in a 6-hydroxydopamine depletion model of Parkinson's disease Vglut3-/- showed normal motor behavior during both waking and sleep cycles suggesting rescue results from more than simply increased dopamine release but also from a form of plasticity. We hypothesize that in the KO mice dopamine depletion triggers maturation of the increased immature spines on direct pathway MSNs and that this normalizes motor behavior across the day/night cycle. We are currently testing our hypothesis by measuring whether there is an increase in the density of mature spines on direct pathway MSNs in Vglut3-/- relative to their WT littermates after DA depletion. We are also recapitulating the elevated DA release in the VGLUT3 KO by targeting an excitatory designer receptor hM3Dq to SNpC DA neurons in Vglut3+/+ mice. This paradigm will allow us test whether a transient increase in DA activity during the waking cycle is sufficient to rescue motor behavior across the day/night cycle in the Parkinson's model a concept that potentially could be applied to patients in early stages of the disease.

First Author: Kase Daisuke (Postdoctoral)	Poster Session: pm
Presenting Author: Kase Daisuke (Postdoctoral)	Location: 25
Mentor/Lab: Robert S. Turner	Category: Neurology & Neurodegenerative Diseases
Department: Department of Neurobiology	

Title: Movement-related activity in the basal ganglia-recipient motor thalamus (VLa) of the parkinsonian macaque

Summary: What causes the symptoms of Parkinson's disease? The principal symptoms of Parkinson's disease slowed movement rigidity and tremor have been recognized for many years. And quite a lot is known about the selective damage to dopamine neurons that is a root cause of those symptoms. Still mysterious however is why a loss of dopamine from a structure deep in the brain leads to this specific cluster of symptoms. We are working on this problem by studying how abnormal neuronal activity spreads from that deep brain structure to impair the operation of brain circuits that control movement. A better understanding of how this kind of malfunction of neural circuits leads to symptoms may lead to enhancements in therapies such as deep brain stimulation for Parkinson's disease.

Abstract: Disordered function of the VLa thalamus is thought to be a critical step in the pathophysiology of motor impairments in Parkinson's disease (PD). For example the traditional "rate model" hypothesizes that elevated discharge rates in efferents from the parkinsonian basal ganglia cause excessive inhibition of VLa neurons which may be evidenced by reduced baseline discharge rates and/or reduced magnitude of movement-related increases in activity. Little information is available however on how the activity of VLa neurons is altered in the parkinsonian state. To address this gap in knowledge we sampled single unit extracellular activities from the VLa before and after (n=99 and 96 units respectively) the induction of hemiparkinsonism by intracarotid MPTP administration in one macaque monkey. The animal performed a simple choice reaction time reaching task for food reward. The animal was able to perform the task throughout the month's-long recording period following MPTP but with markedly prolonged and more variable reaction times and movement durations (reaction time: 248±35 ms vs. 457±215 ms movement durations: 244±35 ms vs. 845±333 ms means±SEM pre- vs. post-MPTP respectively; p<0.01 for both K-S test). The baseline firing rates of VLa neurons sampled during attentive rest while the animal waited for the task's "go" stimulus were not altered by MPTP (14.0±1.2 Hz pre- vs. 12.0±1.1 Hz post-MPTP respectively; p>0.05 K-S test). Large fractions of neurons changed firing rates around the time of reach onset (92% and 83% of neurons pre- and post-MPTP; p=0.07 χ2-test) with increased firing as the earliest change in 79% (pre-MPTP) and 61% (post-MPTP) of these cells and decreased firing as the earliest change in the remainder. This shift following MPTP toward early movement-related decreases in firing was significant (p=0.01; χ2-test). In addition the magnitudes of movement-related increases in firing were reduced markedly following MPTP (20.9±2.2 Hz pre- vs. 5.0±0.7 Hz post-MPTP; p<0.01 K-S test) whereas the magnitude of decreases did not differ (6.2±0.8 Hz pre- vs. 6.1±1.0 Hz post-MPTP). Finally peri-movement activity began earlier relative to movement onset following MPTP (96.5±14.2 ms pre- vs. 176.8±19.9 ms post-MPTP respectively; p<0.01 K-S test). The shift in timing was similar for increases and decreases in firing. The observed MPTP-induced reductions in the prevalence and magnitude of movement-related increases in activity lend partial support for the traditional rate model of PD pathophysiology.

First Author: Daniela Leronni (Faculty)	Poster Session: pm
Presenting Author: Daniela Leronni (Faculty)	Location: 26
Mentor/Lab: Friedlander	Category: Neurology & Neurodegenerative Diseases
Department: Neurological Surgery	

Title: Melatonin Synthesis Enzyme is Misregulated in Huntington's Disease Model

Summary: HD is an autosomal-dominant chronic neurodegenerative disease due to an extended polyQ repeat in the huntingtin (HTT) protein. Mutant HTT (mHTT) protein localizes in brain mitochondria and interferes with the inner membrane mitochondrial importing complex. Mitochondria import defect precedes overt symptoms onset in R6/2 mice suggesting it is an early disease mediating event. Melatonin is a potent endogenous free radical scavenger and it is deficient in humans with HD. The cause and consequence of melatonin deficiency in HD are unknown. Our hypothesis is that AANAT the melatonin synthesis rate-limiting enzyme is actively transported across the mitochondrial membrane and that this transport is disrupted in neurons expressing mHTT.

Abstract: Melatonin is a well-known hormone secreted by the pineal gland and it is involved in circadian regulation. This hormone has several other important functions in the organism and it is shown to be neuroprotective in many neurodegenerative diseases. Melatonin can exert anti-apoptotic effects mainly targeting mitochondria but it can also enhance cell survival pathways leading to cell rescue. In some neurodegenerative diseases for example Huntington's disease (HD) melatonin plasma level is decreased. However circulating melatonin levels regulated by pineal gland activity do not reflect neuronal melatonin levels and the mechanisms for making and maintaining melatonin in neurons is unknown. High levels of melatonin have been found in mitochondria but little is known about the transport of melatonin inside the mitochondria. Our preliminary data show that Arylalkylamine N-acetyltransferase (AANAT) the rate-limiting enzyme in the production of melatonin is in the mitochondria. AANAT must actively be transported from the cytosol across the mitochondrial membranes a process known to be disrupted in HD patients. HD is an autosomal-dominant chronic neurodegenerative disease due to an extended polyQ repeat in the huntingtin (HTT) protein. Recently our group showed that mutant HTT (mHTT) protein localizes in brain mitochondria and interferes with the inner membrane mitochondrial importing complex thus inhibiting mitochondrial protein import. Our data demonstrated that the mitochondrial import defect precedes overt symptoms onset in R6/2 mice suggesting it is an early disease mediating event. Our hypothesis is that AANAT is actively transported across the mitochondrial membrane and that this transport is disrupted in neurons expressing mHTT. The consequences of defective import of AANAT would be decreased melatonin level which could make HD neurons more vulnerable to stress contributing to the pathology of HD disease. How AANAT import is effected in HD mitochondria will provide important insights for future studies to investigate dysregulation of neuronal melatonin synthesis in HD.

First Author: Svitlana Yablonska (Postdoctoral)	Poster Session: pm
Presenting Author: Svitlana Yablonska (Postdoctoral)	Location: 27
Mentor/Lab: Robert M. Friedlander	Category: Neurology & Neurodegenerative Diseases
Department: Neurological Surgery	
Title: Disruption of mitochondrial proteostasis in Huntington disease	
Summary: Mutant huntingtin cause protein disbalance in brain mitochondria of HD patients.	

Abstract: Growing evidence indicates that mitochondria play an important role in the pathogenesis of neurodegenerative diseases including Huntington's disease (HD). The majority of mitochondrial proteins are encoded in the nucleus and imported into mitochondria through pore complexes of translocases of mitochondrial membranes (TOM40 TIM23 TIM22). Mutant huntingtin (mHTT) the causative gene in Huntington's Disease associates with the translocase of mitochondrial inner membrane (TIM23) complex interfering with its normal function [Yano 2014]. To determine the biological consequences of this association we quantified the levels of specific mitochondrial proteins in postmortem frozen human cortex tissue of HD grade 4 patients. We found decreased amounts of the matrix and inner membrane bound proteins that should have been imported through TIM23 complex. Multi-span proteins of inner membrane that are imported using the TIM22 pathway do not change nor do multi-span proteins in the outer mitochondrial membrane. Therefore the association of mHTT with the TIM23 import pathways disturbs mitochondrial proteostasis of specific proteins and may lead to neuronal death in HD pathogenesis.

First Author: Nicole Czachowski (Graduate)	Poster Session: pm
Presenting Author: Nicole Czachowski (Graduate)	Location: 28
Mentor/Lab: Dr. Yijen Wu	Category: Neurology & Neurodegenerative Diseases
Department: Developmental Biology	

Title: MRI Investigation of CDKL5 in Mutant Mouse Models

Summary: CDKL5 is a rare genetic disorder with symptoms that affect neurodevelopment in children. In order to uncover the mechanisms of the disorder mutant mice models were imaged analyzed and compared to unaffected mice revealing significant volume disparities in various brain regions and differing proportions of brain axes. These results suggest that CDKL5 may affect the development of specific brain regions leading to poor patient outcomes.

Abstract: Introduction: CDKL5 is a rare X-linked genetic disorder that entails of a mutation in the CDKL5 gene located on Xp22.13 which codes for the protein cyclin-dependent kinase-like 5. Symptoms of CDKL5 include epileptic encephalopathy arising prior to 3 months of age muscular hypotonia and severe developmental delay. CDKL5 is often associated with Rett Syndrome despite being a separate entity due to similar symptoms and outcomes. Little is known about the etiology or specific neurodevelopmental problems associated with CDKL5. The objective of this study is to elucidate the neurodevelopmental abnormalities associated with CDKL5 in order to better understand the neurological outcomes of the disorder. Methods: Animal model: Genetically modified mice with a mutation in the CDKL5 gene were used in comparison to wild-type (WT) control littermates. The mice were divided into three groups based on age each containing WT and CDKL5 mice. The hemizygous CDKL5 mutants and WT controls were then analyzed using MRI technology. Brain MRI analysis: Multi-modal magnetic resonance imaging (MRI) was utilized to anatomically analyze the mouse brains. Multi-slice 2D T2 WT (RARE8 78 x78 matrix 0.55 mm SLTH) and 3D Heavy T2WT (RARE10 49 x 52 x 52 matrix) were used to acquire gray matter and cerebrospinal imaging respectively. The multi-slice 2D MRI images underwent volumetric and bi-planar analysis and the 3D images underwent volumetric analysis only. Results: Volumetric analysis showed a statistically significant difference in volumes of the cerebral hemisphere corpus callosum cortex subcortex aqueduct and cerebrospinal fluid between the mutant and WT mice in all groups. Bi-planar analysis revealed a statistically significant difference in the ratio of the anterior-posterior and dorsal-ventral axes between the mutant and WT mice. Conclusion: Our results suggest that CDKL5 may cause brain dysplasia in the cerebral hemisphere corpus callosum cortex subcortex and aqueduct as well as abnormal cerebrospinal fluid and axes proportions. These findings suggest that the poor neurological outcomes of CDKL5 patients may be associated with deviations in these brain regions.

First Author: Scott Ginebaugh (Graduate)	Poster Session: pm
Presenting Author: Scott Ginebaugh (Graduate)	Location: 29
Mentor/Lab: Stephen D. Meriney	Category: Neurology & Neurodegenerative Diseases
Department: Neuroscience	

Title: A novel computational model for the development of a new therapeutic approach for Lambert-Eaton myasthenic syndrome

Summary: This research improves our understanding of and examines potential treatments for the disease Lambert-Eaton Myasthenic Syndrome which causes severe muscle weakness. We essentially built the part of the body which is effected by this disease called the neuromuscular junction in a supercomputer which allows us to examine this disease at levels which are not feasible under the microscope or in the laboratory. After building or model in the computer we will use it to estimate the proper dosage of drugs needed to effectively treat this disease which will not only allow us to learn more about the neuromuscular junction and help facilitate the development of treatment for Lambert-Eaton Myasthenic Syndrome but will also help develop a powerful new tool in the drug development process which can be applied to a variety of diseases and conditions.

Abstract: The neuromuscular junction is a reliable synapse in which reliability is derived from the summed activity of numerous unreliable elements each consisting of a synaptic vesicle and associated voltage gated calcium channels (VGCCs). Lambert-Eaton myasthenic syndrome (LEMS) is an autoimmune disease that reduces reliability leading to muscle weakness. This weakness is due to an autoantibody-mediated removal of some of the VGCCs that are critical for transmitter release an upregulation of other VGCC types and a disruption in organization of these VGCCs. LEMS patients are currently managed using a potassium channel blocker (DAP) that broadens the presynaptic action potential. However DAP provides only modest symptomatic relief for LEMS patients. We have previously reported the development of a novel first-in-class Cav2 gating modifier (GV-58) which prolongs channel deactivation effectively increasing calcium flux during an action potential by stabilizing the open state of the channel. We hypothesize that a combination of DAP plus our calcium gating modifier would work synergistically to provide a stronger and more complete relief of neuromuscular weakness. We have built an MCell computational model of the presynaptic neuromuscular active zone to examine the structure-function relationship of the healthy and LEMS disease state neuromuscular junctions. This validated model not only provides us with information about the presynaptic terminal but also allows us to computationally explore various combinations of DAP and GV-58 and study the spatio-temporal dynamics of presynaptic calcium influx and the subsequent impact on transmitter release. The ability to examine the combination of these drugs in silico is particularly important due to the difficulty of creating LEMS model mice. Within MCell we modeled DAP effects by increasing the amplitude (5-10%) and prolonging the decay time (5-15%) of the presynaptic action potential. To model the effects of GV-58 we edited our calcium channel gating scheme to include drug bound states with kinetic rates that resulted in modeled calcium current that matched our patch clamp recordings of calcium current modulation. Then we used these two modifications in both control and LEMS model active zone architecture to evaluate the effects on transmitter release. Our MCell model provides new information on the organization of the transmitter release site and gives us dose-response details for the synergistic effect of GV-58 and DAP which will help facilitate the design of pre-clinical experiments on LEMS model mice.

First Author: Steven Wellman (Graduate)	Poster Session: pm
Presenting Author: Steven Wellman (Graduate)	Location: 30
Mentor/Lab: Takashi Kozai	Category: Neurology & Neurodegenerative Diseases
Department: Bioengineering	

Title: Two-photon imaging reveals processes extension and cell body migration of reactive NG2 glia during acute brain injury

Summary: Recent studies suggest there are other immune cells besides just microglia and astrocytes involved in the development of a glial scar after injury. A critical function of NG2 glia which is to maintain neuronal health and physiology through the formation of synapses with neurons may be compromised after an insult to the brain. Using real time imaging techniques we observed NG2 glia respond to brain implant injury by changes in cell morphology extension of cellular processes and migration of cell bodies toward the lesion site similar to microglia yet at a slower rate indicating novel features of scar development after injury.

Abstract: Activation of microglia and astrocytes and their contribution to neuronal loss are historically the focus of investigations into scar tissue formation after brain injury. However recent studies have implicated other effectors in the progression of reactive gliosis. NG2 glia which arise from the oligodendrocyte lineage during development are widely distributed across the adult brain and exist as a separate glial entity with distinctive characteristics. Known also as oligodendrocyte precursor cells they are responsible for differentiating into myelinating oligodendrocytes in normal CNS physiology and after incidences of demyelination. Unique to glial cells NG2 glia form functional synapses on neurons with the ability to influence neuronal viability through secretion of neurotrophic factors and modulation of neuronal networks. Therefore NG2 glia may display alternative behavior under pathological conditions that could potentially be detrimental to brain tissue health. After injury NG2 expression is known to increase following migration and proliferation of NG2-expressing cells around lesion sites. Due to their intrinsic potential to differentiate into astrocytes and express axon-growth inhibitory molecules NG2 glia have been implicated in the formation of the glial scar. Here we use in vivo two-photon microscopy to characterize the initial NG2 glia scar formation around brain implant injuries in the cortex through changes in cell shape transforming from an inactive ramified state to a transitional morphology. Similar to microglia NG2 glia are seen extending cellular protrusions and migrating towards the surface of the electrode. However unlike microglia cells who respond immediately on the order of minutes to electrode insertion NG2 glia do not extend processes or migrate cell bodies until hours post-insertion. This delay in cell response between microglia and NG2 cells may imply unique possibly chemotactic cell-cell interactions between glia in the reactive tissue response after injury. Fully comprehending the role of NG2 glia in the disease state and their divergence from normal physiological function can offer previously unknown insights into the inflammatory tissue reaction after brain injury and potentially foster novel strategies towards attenuating those responses.

First Author: Lily Francis (Graduate)	Poster Session: pm
Presenting Author: Lily Francis (Graduate)	Location: 31
Mentor/Lab: Chu Lab/ Charleen Chu	Category: Neurology & Neurodegenerative Diseases
Department: Neuropathology/ Human Genetics	

Title: Neuropathology of POLG-related mitochondrial diseases in patient-derived iPSC-neurons

Summary: We describe the use of stem cell derived neurons from patients as a model for the study of Neurodegenerative diseases.

Abstract: DNA polymerase gamma (Polg) is responsible for mitochondrial DNA (mtDNA) replication and repair. Mutations in POLG the gene encoding the catalytic subunit of Polg result in a set of clinical syndromes characterized by mtDNA depletion in affected tissues with variable organ involvement and severity. The brain and neuromuscular system are the most commonly affected organs with intractable seizures developmental delay dementia ataxia liver failure axonopathies myopathy and ophthalmoplegia comprising major symptoms. Treatment for POLG-related disorders remains mostly supportive with the majority of patients progressing to severe disability and death within a few years of diagnosis. Therefore a better understanding of disease mechanisms in the affected cell types is needed to illuminate new therapeutic options for these devastating diseases that typically affect children and teenagers. Most patients with POLG mutations are compound heterozygotes bearing a different mutation in each allele. Here we describe our work studying cortical neurons differentiated from two new patient-derived models of POLG-related mitochondrial diseases (POLG1 and POLG3). Fibroblasts from diagnostic skin biopsies were reprogrammed into induced pluripotent stem cells (iPSCs) and mutation status confirmed by DNA sequencing. While the patient-derived iPSCs did not show mtDNA depletion relative to control iPSCs both POLG1 and POLG3 failed to undergo the dramatic increase in mtDNA content observed in control lines upon differentiation to cortical neurons. Neurons differentiated from patient iPSCs exhibited simplification and shortening of the neuritic arbor with multiple abnormal neuritic swellings. POLG1 and POLG3 also exhibited abnormal mitochondrial ultrastructure by electron microscopy with accumulation of autophagic vacuoles and altered neuritic trafficking of lysosomes. Ongoing studies are aimed at characterizing mitochondrial function and dynamics in somatic and neuritic compartments and use of gene editing or other strategies to reverse these pathological neuronal phenotypes.

First Author: Jacob Mann (Graduate)	Poster Session: pm
Presenting Author: Jacob Mann (Graduate)	Location: 32
Mentor/Lab: Donnelly	Category: Neurology & Neurodegenerative Diseases
Department: Neurobiology	

Title: Optogenetic Induction of TDP-43 Proteinopathy

Summary: Aggregation of various disease-linked proteins is a common pathological process experienced in neurodegenerative diseases such as Alzheimer's Disease Amyotrophic Lateral Sclerosis and Parkinson's Disease among others. However experimentally controlling this process of protein aggregation has been historically problematic. Here we show a new model of protein aggregation by using light-responsive proteins isolated from plants that allows for a previously unachievable level of spatial and temporal control.

Abstract: Over the last twenty years mutations in over 35 different genes have been linked to the development of familial forms of ALS (fALS); however fALS only accounts for roughly 10% of all ALS cases. The remaining 90% of patients suffer from sporadic ALS (sALS) with no family history of disease and unknown causes of pathogenesis. Regardless of all this genetic and pathogenic complexity remarkably nearly every single ALS patient (~97%) shares a common neuropathology in the form of cytoplasmic aggregates of a protein called TAR DNA-binding protein of 43 kDa (TDP-43) found in degenerating regions of the nervous system. Current cellular models of this neurodegenerative proteinopathy often rely on the overexpression of disease-linked mutant proteins to induce pathological protein aggregation. However mutations in the TARDBP gene only account for ~1% of sporadic (sALS) and 4% of familial ALS (fALS) cases. The vast majority of patients do not harbor mutations in this gene yet still experience TDP-43 aggregation. Similarly rodent models of ALS produced from the overexpression of these mutant proteins have been historically unreliable and often fail to generate TDP-43-positive inclusions. Here we present a novel optogenetic-based technique to induce pathological aggregation of TDP-43 with a previously unachievable level of spatial and temporal control. Using this approach we show the tunable oligomerization and aggregation of TDP-43 and disease-related truncations of the protein in response to varying light stimulation paradigms. These induced TDP-43 aggregates share similar pathological characteristics with TDP-43 inclusions observed in ALS patient tissue and also appear to result in endogenous TDP-43 loss-of-function mechanisms that have been previously implicated in disease progression. Utilizing this technique we have uncovered perturbed oligomerization dynamics due to fALS-linked mutations in TDP-43 that may underlie the enhanced aggregation properties and neurotoxicity of these mutated proteins. We have also begun to identify novel pathway modulators of TDP-43 aggregation that may play a role in the development of both fALS and sALS. This technique can be applied to a number of different disorders will allow for more precise temporal and spatial control over protein aggregation than has been previously possible. Additionally the ability to reliably induce protein aggregation with light alone will allow for in-depth investigations into the effects of these pathological aggregates on various cellular pathways and downstream pathological processes.

First Author: Abhishek Jauhari (Postdoctoral)	Poster Session: pm
Presenting Author: Abhishek Jauhari (Postdoctoral)	Location: 33
Mentor/Lab: Dr Robert Freidlander	Category: Neurology & Neurodegenerative Diseases
Department: Department of Neurological surgery	

Title: Absence of endogenous melatonin induced immune response mediated synaptic degeneration in differentiated neurons

Summary: AANAT KO leads to absence of endogenous melatonin which in turn to results in accumulation of ROS and MMP loss of mitochondria. Elevated ROS and hypopolarized mitochondria activate immune response which results in synaptic and neuritic degeneration and finally neuronal cell death.

Abstract: Absence of endogenous melatonin induced immune response mediated synaptic degeneration in differentiated neurons Abhishek Jauhari Sergei Baranov Svitlana Yablonska Diane L Carlisle and Robert Friedlander* Department of Neurological surgery University of Pittsburgh Medical center Pittsburgh PA USA *Corresponding Author Melatonin is an endogenously occurring free radical scavenger and well documented in neuroprotection as it reduced the loss of neurons under pathophysiological conditions. Therefore to test whether endogenous melatonin is involved in regulation of neuronal development and neurodegeneration we developed CRISPR/CAS9 mediated Arylalkylamine N-acetyltransferase (AANAT) knockout (KO) N2A cells. AANAT is a rate limiting enzyme in the synthesis of melatonin from serotonin. Wild type (WT) and AANAT KO N2A cells were differentiated into mature neurons by the exposure of retinoic acid. Our studies have demonstrated that differentiated AANAT KO cells have elevated reactive oxygen species (ROS) and significant loss in mitochondrial membrane potential (MMP) in comparison to their wild type differentiated N2A cells. Further qPCR studies has shown that differentiated AANAT KO cells have increased level of inflammatory markers (IL6 TNFα IFNA IFNB). In addition our studies has revealed that differentiated AANAT KO cells have lower number of synapses decreased average length of neurites and neurite numbers. Interestingly when AANAT KO cells were treated with melatonin during differentiation the synaptic degeneration neuritic length neuritic numbers MMP and ROS were rescued similar to their WT differentiated N2A cells. In addition AANAT KO differentiated N2A cells have shown the decrease in level of inflammatory markers when grown with melatonin. In conclusion AANAT KO leads to absence of endogenous melatonin which in turn to results in accumulation of ROS and MMP loss of mitochondria. Elevated ROS and hypopolarized mitochondria activate immune response which results in synaptic and neuritic degeneration and finally neuronal cell death.

First Author: Fen Pei (Graduate)	Poster Session: pm
Presenting Author: Fen Pei (Graduate)	Location: 34
Mentor/Lab: Lansing Taylor	Category: Neurology & Neurodegenerative Diseases
Department: Computational biology department	

Title: Connecting Neuronal Cell Protective Pathways and Drug Combinations in a Huntington's Disease Model through the Application of Quantitative Systems Pharmacology

Summary: Through the application of a chemogenomics platform we investigated the protective effects of small molecule probes and probe combinations for HD disease model Computational analysis of these probes revealed a convergence of pathways indicating activation of PKA.

Abstract: Quantitative Systems Pharmacology (QSP) is a drug discovery approach that integrates computational and experimental methods in an iterative way to gain a comprehensive unbiased understanding of disease processes to inform effective therapeutic strategies. We report the implementation of QSP to Huntington's Disease with the application of a chemogenomics platform to identify strategies to protect neuronal cells from mutant Huntingtin induced death. Using the STHdhQ111 cell model we investigated the protective effects of small molecule probes having diverse canonical modes-of-action to infer pathways of neuronal cell protection connected to drug mechanism. Thirty-two mechanistically diverse protective probes were identified most of which showed less than 50% efficacy. Specific combinations of these probes were synergistic in enhancing efficacy. Computational analysis of these probes revealed a convergence of pathways indicating activation of PKA. Analysis of phospho-PKA levels showed lower levels in the cytoplasm of STHdhQ111 cells compared to the wild type STHdhQ7 cells and these levels were increased by several of the protective compounds. In addition the PKA inhibitor H89 at pharmacodynamically active non-toxic concentrations inhibited the effects of several protective compounds thereby supporting the hypothesis that these protective compounds may be working in part through activation of the PKA network. The systems-level studies described here can be broadly applied to any discovery strategy involving small molecule modulation of disease phenotype.

First Author: Sergei Baranov (Postdoctoral)	Poster Session: pm
Presenting Author: Sergei Baranov (Postdoctoral)	Location: 35
Mentor/Lab: Friedlander R.F.	Category: Neurology & Neurodegenerative Diseases
Department: Neurological Surgery	

Title: Mitochondria controlled local caspase-3 activation in neuronal processes. Single-cell analysis

Summary: Under the stress mitochondrial failure in distal neuronal compartments induce activation of caspase-3 in neuronal processes

Abstract: Human studies reveal synaptic dysfunction decades before predicted clinical diagnosis in neurodegenerative diseases. Loss of dendrites and synapses requires activation of apoptotic terminal protease caspase-3 but does not always lead to immediate cell death. Loss of synapses is a characteristic of Alzheimer's and Huntington's diseases. Activation of caspase-3 is mitochondria dependent. Damage to mitochondria results in release of cytochrome c and activation of caspase-3. We hypothesized that Huntington's disease associated synaptic loss among other factors caused by mitochondria- dependent local caspase-3 activation without immediate cell death. Using single cell analysis approach we assessed modulation of local caspase-3 activity in the primary neurons from mouse model of Huntington's disease. We found that activation of caspase-3 in axo-dendritic neuronal compartments was associated with mitochondrial depolarization under excitotoxic conditions. We showed that found elevated activity of caspase-3 in distal compartments of neurons of Huntington's disease model was correlated with a decreased mitochondrial membrane potential increased level of oxidized/damaged mitochondrial protein content and an increased production of reactive oxygen species by mitochondria found in the same compartments. We explained our data in the framework of mitochondria-dependent cytochrome c-associated activation of caspase-3 in distal neuronal compartments where mitochondria are more vulnerable to stress associated with the neurological disorder.

First Author: Patricia B. de la Tremblaye (Postdoctoral)	Poster Session: pm
Presenting Author: Patricia B. de la Tremblaye (Postdoctoral)	Location: 36
Mentor/Lab: Anthony E. Kline	Category: TBI-Concussion
Department: Physical Medicine and Rehabilitation	

Title: Long-term effects of adolescent chronic stress on TBI cognitive and emotional impairments in adult male rats

Summary: The most common neuropsychiatric consequence of TBI is depression. Early stress exposure has been recognized as an important mechanism for neuropsychiatric disorders in adulthood. In rodents as in humans adolescence is a transitional period between child- and adult-hood that is marked by behavioral changes heightened brain development and cognitive maturation. Therefore exposure to adverse environmental conditions during this sensitive period of development could influence TBI psychiatric outcomes. Therefore the current study examines weather repeated stress during adolescence will result in deleterious effects on emotional and cognitive functional impairments in rats subjected to a TBI as adults. Understanding the impact of environmental factors underlying post concussive symptoms will help develop effective preventive and therapeutic strategies for TBI patients.

Abstract: Exposure to early life stress has lasting effects on behavior and brain function due to dynamic plasticity occurring in the developing adolescent brain. However it is yet to be determined how stress exposure in this developmental period influences functional recovery post traumatic brain injury (TBI) induced later in life. Thus the goal of this study was to test the hypothesis that stress in adolescence would confer deleterious effects on behavioral impairments post TBI in adulthood. Adolescent male Sprague-Dawley rats (n=40) were exposed to 4 weeks (postnatal day PND 30-60) of chronic unpredictable stressors (CUS) or no stress and after a 1-month resting period (PND 60-90) were anesthetized and received a cortical impact of moderate severity (2.8 mm tissue deformation at 4m/s) or sham injury. After one week of recovery anxiety-like behavior in the open field test (OFT) and elevated plus maze (EPM) and cognitive performance in the novel object recognition (NOR) task and Morris water maze (MWM) were measured. Brains were collected 25 days after TBI for histological analysis. Preliminary results show increased time spent in the anxiogenic zones of the OFT and EPM and improved NOR memory after a 24 h delay in addition to reduced time to reach the platform in the MWM for CUS groups compared to no-stress control groups although TBI rats remained significantly more anxious and cognitively impaired compared to sham controls. These results suggest that aversive environmental conditions in adolescence induces adaptive behavioral responses in TBI rats albeit without leading to full functional recovery.

First Author: Daniel Charek (Postdoctoral)	Poster Session: pm
Presenting Author: Daniel Charek (Postdoctoral)	Location: 37
Mentor/Lab: Anthony Kontos PhD	Category: TBI-Concussion
Department: Department of Orthopaedic Surgery	

Title: Predicting Patients with Vestibular Clinical Profiles following Concussion

Summary: Concussions may involve different clinical profiles and this study sought to determine which factors best predict patients with a vestibular profile which is associated with poor clinical outcomes and recovery times. Of relevant factors included in a statistical model for predicting participants with vestibular clinical profiles a history of motion sickness and combined nausea dizziness and fatigue symptoms were positive predictors. These factors should be considered by clinicians when evaluating patients to facilitate identification of the vestibular profile so that appropriate targeted treatments can be prescribed.

Abstract: Objective: Concussions may involve different clinical subtypes or profiles including cognitive anxiety/mood migraine oculomotor and vestibular (Collins Kontos Reynolds et al. 2014). Early identification of clinical profiles is critical to inform effective and timely treatments. The vestibular clinical profile is associated with poor clinical outcomes and longer recovery times (Corwin Wiebe Zonfrillo et al. 2015; Lau Kontos Collins et al. 2011). The aim of this study was to determine which factors best predict patients with a vestibular clinical profile. Methods: Participants included 50 adolescent patients aged 12-20 years with a diagnosed sport-related concussion. Participants were divided into either: 1) vestibular or 2) other clinical profile groups based on positive findings on a vestibular screening exam clinical evaluation and subsequent follow-up testing. A logistic regression (LR) model was used to predict participants with vestibular profiles. Predictors included: gender; age; history of motion sickness migraine and concussion; dizziness at time of injury; computerized neurocognitive scores; clinical balance performance; and specific symptoms. Results: The LR was significant (p<.001 Nagelkerke R2=.51) with history of motion sickness (p=.02) and combined nausea dizziness and fatigue symptoms (p=.002) as positive predictors of the vestibular profile. Sensitivity for the model was 81.0% and specificity was 85.2%. Conclusion: A history of motion sickness and higher reported nausea dizziness and fatigue are useful predictors of patients with vestibular clinical profiles. Clinicians should focus on these factors when evaluating patients to better identify those with vestibular profiles to allow for more effective precision treatments.

First Author: Natalie Sandel (Postdoctoral)	Poster Session: pm
Presenting Author: Natalie Sandel (Postdoctoral)	Location: 38
Mentor/Lab: Anthony Kontos PhD	Category: TBI-Concussion
Department: Orthopaedic Surgery	

Title: Comparing near point of convergence distance in concussed adolescents and healthy controls

Summary: Adolescent athletes evaluated within 10 days of their concussion demonstrate a convergence insufficiency a reduced ability for the eyes to team together upon near vision relative to healthy controls. Convergence appears to return back to normal when concussed athletes are cleared to return back to sports.

Abstract: Near vision oculomotor dysfunction such as an accommodation or convergence insufficiency is common after brain injury. Nearly 40% of athletes with a sports-related concussion exhibit a convergence insufficiency in which there is a reduced ability for the eyes to team together upon near vision (Pearce et al. 2015). A convergence insufficiency can cause several symptoms including blurred or double vision headache and difficulty with reading or computer work. Despite evidence of oculomotor deficits after brain injury limited research has explored whether these posttraumatic vision changes remit after a concussion. The goal of the current study was to compare near-point convergence (NPC) distance within concussed individuals at the time of initial visit compared to their NPC distance at the time of clearance back to sports based on international criteria (McCrory et al. 2012) relative to a group of healthy controls. Participants were aged 12 to 20 years old (M=15.13 SD=2.05). A total of 39 concussed participants (53.8% male) were matched closely to healthy controls (N=28 64.3% male). Concussed participants were diagnosed with a concussion by a neuropsychologist at their initial visit within 10 days of their injury (M=6.51 SD=2.55) and serially assessed across subsequent follow-up visits. NPC was measured in healthy controls at only one time point. Among concussed individuals 59.8% were formally cleared by their fourth visit (Mdn=22 days post injury Range=10-193 days). All participants underwent Vestibular/Ocular Motor Scoring (VOMS) screening including NPC measurement. An average NPC measurement greater than 5cm was considered abnormal (Mucha et al. 2014; Scheiman et al. 2003). Independent t-tests were conducted to determine if concussed athletes differed from controls at initial visit and time of clearance. A paired samples t-test compared NPC measurements of concussed athletes at initial visit and time of clearance. Results of an independent t-test comparing NPC at initial visit in concussed (M=6.88cm SD= 10.05cm) versus controls (M=1.61cm SD= 2.54cm) was significant $t(43.25)=3.10$ $p=.003$ with concussed participants demonstrating poorer NPC as a group. In the control group 14.3% demonstrated an abnormal NPC while in the concussed group 36.8% demonstrated an abnormal NPC. A paired-samples t-test for comparison of the concussed participants' NPC at initial visit (M=6.34 SD=10.36) versus at time of clearance (M=1.77 SD=2.82) yielded a significant difference $t(21)=2.23$ $p=.04$ with greater NPC at the initial visit. Lastly an independent t-test for NPC between concussed participants at their clearance visit (M=1.81cm SD= 2.76cm) versus controls (M=1.61 SD=2.54cm) was non-significant $t(49)=0.27$ $p=.79$ indicating no difference in NPC between individuals cleared from their concussion and healthy controls. Individuals with a concussion demonstrate a significantly worse near point of convergence initially after injury relative to healthy controls. Abnormal near point of convergence after concussion appears to return to normal over time and/or with treatment. This study was limited by the attrition of participants who did not return for follow-up evaluation and a lack of repeated measurement for the control group.

First Author: Jordan Brooks (Graduate)	Poster Session: pm
Presenting Author: Jordan Brooks (Graduate)	Location: 39
Mentor/Lab: Ava Puccio	Category: TBI-Concussion
Department: Department of Neurosurgery	

Title: Differential CSF Cytokine Profile of Patients with Post-Traumatic Hydrocephalus

Summary: This project compares the inflammatory response of severe traumatic brain injury patients who developed post-traumatic hydrocephalus to severe traumatic brain injury patients who did not develop post-traumatic hydrocephalus. The goal of the project was to assess whether the inflammatory reaction ensuing traumatic brain injury influenced the development of post-traumatic hydrocephalus. Ultimately this project will give us a better understanding of the cause of post-traumatic hydrocephalus which has been associated with worst outcomes in this patient population to ultimately allow for intervention.

Abstract: Post-traumatic hydrocephalus (PTH) is a secondary neurological insult resulting in the derangement of cerebrospinal fluid (CSF) dynamics ensuing moderate to severe traumatic brain injury (sTBI). Given the high risk of clinical deterioration and documented worse outcomes the identification of biomarkers indicating the onset of PTH is imperative to allow early clinical detection and improve neurological outcomes in afflicted patients. This study examined CSF cytokine profile with PTH to elucidate the pathogenesis and aide in the early diagnosis of PTH. We conducted a matched case-control study on 50 patients who sustained a sTBI at a level 1 Trauma facility from 2002-2015. All patients were treated with five days of continuous CSF drainage via an extraventricular drain. CSF research samples was collected on post-trauma days 1 3 and 5. Patients who incurred CNS infection or died within 6 months were excluded. 25 patients who incurred PTH were matched by age sex and initial Glasgow Coma Scale with 25 patients who did not incur PTH. The CSF concentrations of 36 different inflammatory markers were analyzed via a Luminex Array Scanner. There were no PTH differences detected between the groups in CSF RBC WBC. Across all time points IL-15 ($p=0.007$) IL-5 ($p=0.038$) and CX3CL1 ($p=0.031$) were significantly lower among PTH patients. CCL4 was significantly higher in the PTH group ($p=0.029$). IL-2 levels increased at a significantly slower rate in patients with PTH ($p=0.037$). No other statistically significant differences were found in any other of the cytokines assayed. Overall our data suggests potential differences in the immune responses in patients who develop PTH which may impede the clearing of debris following sTBI. Lower levels of IL-15 and IL-5 suggest the decreased recruitment and proliferation of natural killer cells T-cells B-cells and eosinophils in PTH patients. High levels of CCL4 may indicate a more macrophage rich environment. A slower increase in IL-2 may be indicative of a global impairment of immune function particularly T-cell function and B-cell differentiation.

First Author: Aaron Sinnott (Graduate)	Poster Session: pm
Presenting Author: Aaron Sinnott (Graduate)	Location: 40
Mentor/Lab: Neuromuscular Research Lab/Christopher Connaboy Anthony Kontos	Category: TBI-Concussion
Department: Sports Medicine and Nutrition	

Title: Do Changes in Symptom Burden Affect Clinical Outcomes following Concussion?

Summary: Post-concussion symptoms are hallmark signs of injury and injury prognosis. The current study investigated clinical measures with symptom complaints after injury.

Abstract: Objective: Initial symptom burden is an important factor to consider when evaluating concussed patients as it is prognostic of poor clinical outcomes. However researchers have yet to examine the role of changes in symptom burden in relation to clinical outcomes following concussion. The aim of the current study was to compare neurocognitive performance across recovery in adolescents and adults who did not improve in symptoms to those who did improve. Design: The study employed a prospective repeated measures design and involved clinical data collected from a concussion testing program between 2009 and 2017. A total of 243 (96F/147M) adolescents and adults aged 14-29 years within 7 days of clinically diagnosed concussions were enrolled in the study. Participants were categorized as either: 1) improved (upper tertile) or 2) not-improved (lower tertile) based on changes in reported symptoms from within 7 days post-injury to within 10 days post-injury. All participants completed the Immediate Post-Concussion Assessment and Testing (ImPACT) and Post-concussion Symptom Scale (PCSS) at baseline 3 and 10 days post-injury. A series of 2 (group) x 3 (time) ANOVAs with Bonferroni correction were conducted for verbal and visual memory visual motor processing speed and reaction time. Results: 162/243 (30.8%) eligible participants were included in the sample. 81 participants (45.7% 37F) were categorized as improved and 81 (25.9% 21F) were categorized as not-improved. Results supported group X time interactions for verbal memory F (2 159) =11.80 p<.001 η2=.13 visual motor speed F (2 159) = 8.710 p<.001 η2=.10; and reaction time F (2159) =12.80 p<.001 η2=.14. Participants in the improved group performed worse than the not improved group at 3 days post-injury on visual and verbal memory and reaction time (p=.001-.003); but not 10 days after injury. As expected there were significant within subjects changes from pre to post-injury across all outcomes (p=.001-.02). There were no differences in outcomes from baseline to 10 days post-injury. Conclusions: Adolescents and adults with large symptom fluctuations perform worse initially but neurocognitive deficits recover compared to those who experience little symptom change after injury. Findings confirm that neurocognitive performance will resolve 10 days after injury despite variations in initial symptom burden.

First Author: Brandon Gillie (Postdoctoral)	Poster Session: pm
Presenting Author: Brandon Gillie (Postdoctoral)	Location: 41
Mentor/Lab: UPMC Sports Medicine Concussion Program	Category: TBI-Concussion
Department: Department of Orthopaedic Surgery	

Title: Association of High-definition Fiber Tracking to Recovery Time and Clinical Outcomes in Adolescents following Concussion

Summary: Advanced neuroimaging techniques including high definition fiber tracking may help to predict recovery from concussion. Adolescents who were slow to recover from concussion showed differences in white matter tracts that were not present among those who recovered quickly.

Abstract: Objective: Findings from conventional imaging techniques such as CT scans and MRI are typically normal following concussion. Evidence of damage to white matter tracts following concussion using diffusion tensor imaging (DTI) though better than convention approaches has been equivocal. There is a need for a better approach to quantify structural damage to white matter following concussion as there are no established markers of brain injury that might identify athletes at risk for prolonged recovery and correlate with clinical findings. One such approach may involve high definition fiber tracking (HDFT). The aim of this study was to examine the association of HDFT white matter tractography metrics at 1-14 days post injury with recovery time and clinical outcomes in concussed adolescent athletes. Methods: Participants included 26 (9F/17M) adolescents aged 15.7 +/- 2.7 years with a diagnosed currently symptomatic concussion. Participants were divided into long (45+ days; n=16) and short (<45 days; n= 10) recovery groups. All participants completed HDFT scans and clinical outcome measures including self-reported post-concussive symptoms computerized neurocognitive testing and vestibular/oculomotor symptoms and impairment within 14 days of injury. Correlations and split-half comparisons of HDFT tract metrics (spread symmetry streamlines) between recovery groups were performed. Correlations adjusted for multiple comparisons were conducted between HDFT metrics and clinical outcomes. Results: Participants with long recovery times had fewer streamlines in the L optic (p=.03) and L thalamic (p=.01) radiations and less variability of streamline length in the R frontal aslant (p=.02) and uncinate (p=.04). They also had longer streamlines in L arcuate (p=.03) and L frontal-occipital fasciculus (p=.04). There were numerous positive correlations among HDFT spread symmetry and streamlines and clinical outcomes (p<.05). Conclusions: HDFT metrics were associated with both recovery time and clinical outcomes. However these associations in this sample of concussion patients at acute/sub-acute post-injury time point differ from those reported in moderate to severe TBI patients at chronic post-injury time points.

First Author: Brenden Tervo-Clemmens (Graduate)	Poster Session: pm
Presenting Author: Brenden Tervo-Clemmens (Graduate)	Location: 42
Mentor/Lab: Laboratory of Neurocognitive Development	Category: Psychiatry
Department: Psychology & Psychiatry	
Title: Brain-based Structure of Psychiatric Comorbidity	
Summary: People with one psychiatric disorder frequently meet diagnostic criteria for another disorder. However little is known about the brain-basis for the co-occurrence of psychiatric disorders. In this project we demonstrate core brain systems associated with cognition and emotion are associated with multiple psychiatric disorders.	
Abstract: Across the lifespan latent variable modeling reveals dimensional higher-order psychopathology factors that account for patterns of comorbidity amongst common psychiatric disorders. However little is known about the structure of psychiatric comorbidity in the brain. To identify neural systems associated with psychiatric comorbidity we utilized resting-state functional magnetic resonance imaging (rsfMRI) data and psychopathology symptom endorsement from 748 subjects of the Philadelphia Neurodevelopment Cohort. Symptom-severity connectivity matrices were created for nine psychiatric disorders and patterns were examined using exploratory factor analysis. Four factors emerged representing general psychopathology (p) fear approach-avoidance and externalizing behavior. Regional expression of these higher-order factors implicated brain-regions associated with transdiagnostic cognitive (DLPFC general psychopathology; ACC externalizing behavior) and affective (amygdala fear; OFC & basal ganglia approach-avoidance) behaviors. Our results suggest common neural systems may contribute to multiple psychiatric disorders highlighting the importance of investigating core psychopathology features in clinical neuroimaging.	

First Author: Chelsea Vadnie (Postdoctoral)	Poster Session: pm
Presenting Author: Chelsea Vadnie (Postdoctoral)	Location: 43
Mentor/Lab: Colleen McClung	Category: Psychiatry
Department: Psychiatry	

Title: Using optogenetics to determine the role of the suprachiasmatic nucleus in mood-like behaviors

Summary: Disruptions in circadian rhythms rhythms that repeat approximately every 24 hours commonly occur in people with mood disorders. The suprachiasmatic nucleus (SCN) in the brain drives and synchronizes circadian rhythms but it is unclear whether perturbing SCN neural activity affects mood. Here we have developed a procedure to study the mood-like effects of delaying or advancing SCN activity in mice.

Abstract: Circadian rhythm disruptions commonly occur in mood disorders. Recent clinical findings suggest that phase delayed rhythms more commonly occur during depressive episodes whereas phase advanced rhythms more frequently occur during manic episodes. The suprachiasmatic nucleus (SCN) synchronizes bodily rhythms with the environment and may underlie the misaligned rhythms observed in mood disorders. Recently disrupting molecular rhythms in the SCN was shown to cause mood-like disturbances in mice suggesting that disrupting SCN neural activity rhythms may affect mood. Thus our goal was to develop a model system to determine if phase-delaying and phase-advancing manipulations of SCN neural activity have differential effects on mood-like behaviors. Channelrhodopsin-2 (ChR2) was genetically introduced into the SCN by crossing mice expressing Cre recombinase in GABAergic neurons with mice expressing Cre-dependent ChR2. Optic fibers were implanted above the SCN and mice were housed in cages equipped with piezoelectric floor sensors to monitor circadian rhythms and sleep. Mice were then placed in constant darkness (DD) to observe their SCN-driven rhythms. Mice subsequently received stimulations (1 h 10 ms pulse width 8 Hz) every three days at times early or late into their active phase to induce phase delays or phase advances respectively. After six stimulation sessions mood-like behaviors were assessed. Stimulating the SCN early in the active phase induced phase delays increasing the period of activity rhythms (24.40 ± 0.06 hr) relative to control mice (24.13 ± 0.06 hr). Stimulating the SCN late in the active phase induced phase advances decreasing the period of activity rhythms (23.55 ± 0.07 hr) relative to controls (23.95 ± 0.02 hr). Thus optogenetic stimulation of GABAergic neurons in the SCN induced phase shifts in circadian activity rhythms that resembled the known effects of light pulses applied in DD. We are currently assessing the effects of the stimulation paradigms on mood-like behaviors. Importantly we have developed a model system to determine the role of SCN-mediated phase shifts of circadian rhythms in mood regulation.

First Author: Maxwell Wang (Graduate)	Poster Session: pm
Presenting Author: Maxwell Wang (Graduate)	Location: 44
Mentor/Lab: Howard Aizenstein MD PhD	Category: Psychiatry
Department: Psychiatry	

Title: Predicting Remission in a Late-Life Depression Treatment Trial

Summary: Identifying an effective depression treatment regimen requires a lengthy trial and error cycle where each drug must be taken for several weeks before a clinician can determine whether the drug was effective. As this cycle continues the patient often spirals further into depression leading to worsening outcomes. Here we present the usage of functional MRI and machine learning towards shortening these several weeks of trial and error down to a 24 hour experiment.

Abstract: Treatment of major depressive disorder typically involves a lengthy trial and error process (around 6-8 weeks in the late-life depression subtype) to identify an effective regimen. This lengthy period delays overall improvement causes patients to drop from care and increases risk of suicide. These patterns are even worse in late-life. However recent work demonstrates that during a venlafaxine (serotonin-norepinephrine reuptake inhibitor) trial significant perturbations in neural functional connectivity occurred rapidly (within 24 hours) following the first dose. In this project we propose an analysis framework to translate these perturbations in functional networks into accurate predictors of clinical remission. Utilizing ten-fold cross-validation and ROC-based metrics we find that our approaches yield significant increases in predictive accuracy over baseline clinical measures such as the Montgomery-Asberg depression rating scale (MADRS). We hope that our model also provides additional insight into the mechanism of venlafaxine within the context of the brain's latent network architecture to motivate possible ways to refine and improve treatment options.

First Author: Kale Edmiston (Postdoctoral)	Poster Session: pm
Presenting Author: Kale Edmiston (Postdoctoral)	Location: 45
Mentor/Lab: Mary Phillips	Category: Psychiatry
Department: Psychiatry	

Title: Predicting quality of life in distressed young adults: Visual cortex and thalamic BOLD signal during reward processing

Summary: In people at risk for depression or anxiety disorders brain activity while waiting to receive a reward predicted their overall quality of life six months later. Parts of the brain related to visual processing were more active among people who had better quality of life later on. This could be related to how visually interesting or noticeable the cue for a future reward is to people with depression and anxiety symptoms; people who have more of a response to reward tend to be functioning better six months later.

Abstract: Study: Identification of neurobiological factors that predict quality of life (QoL) in mood and anxiety disorders could help identify young adults requiring more targeted treatment. Alterations in reward processing are a core component of mood and anxiety disorders. Functional MRI research indicates associations between BOLD during reward processing and mood and anxiety symptoms. However it is unclear how such alterations might predict later QoL. Methods: In this fMRI study twenty-eight young adults (ages 18-25) experiencing psychological distress completed an uncertain reward task in scanner. Participants then returned for a six-month follow-up and completed the Quality of Life Enjoyment and Satisfaction Questionnaire (QLESQ). Correlation between BOLD signal during reward expectancy or BOLD signal during prediction error and QoL as assessed by the change in QLESQ Total Scores at time one and six-month follow-up was modeled. Results: There were significant positive correlations between change in QoL at follow-up and BOLD signal during reward expectancy in the dorsomedial thalamus cuneus and left primary visual cortex such that increased BOLD was associated with improved QoL (p<0.001 uncorrected). There was also a significant positive correlation between QoL at follow-up and BOLD in the left premotor cortex during the prediction error portion of the task. Conclusion: Our findings indicate that enhanced activity of cortico-thalamic regions during reward processing is predictive of later QoL in a distressed sample of young adults. Significance: These findings may help to identify neurobiological features associated with improved outcomes in mood and anxiety disorders potentially leading towards targeted therapeutic interventions.

First Author: Gabrielle Kaplan (Graduate)	Poster Session: pm
Presenting Author: Gabrielle Kaplan (Graduate)	Location: 46
Mentor/Lab: Logan/McClung	Category: Psychiatry
Department: Psychiatry	

Title: Mitochondrial complex I alterations in a mouse model of bipolar mania

Summary: Our studies show that the ClockΔ19 mouse as a model for bipolar mania recapitulates the mitochondrial alterations found in human postmortem tissue and will serve as a model for future studies investigating the direct links between circadian clock machinery cellular metabolism and mitochondrial respiration.

Abstract: Study: A confluence of evidence points towards an underlying dysfunction of mitochondrial complex I in bipolar disorder (BD) which may lead to an increase in oxidative stress and inflammation. The Clock mutant mice (ClockΔ19) which has been shown to display a behavioral repertoire similar to bipolar mania serves as a model in which we can investigate both the circadian control of complex I and potential neuronal mitochondrial dysfunction in the prefrontal cortex a critical structure known to regulate mood and decision making in the brain. Methods: Live mitochondria from WT and ClockΔ19 mouse PFC brain tissue were isolated and separated via density centrifugation. Mitochondria then underwent high-resolution respirometry measures to obtain rate of oxygen consumption. RCR (respiratory control rate) was calculated by dividing State 3 (ADP-stimulated) and State 2 (substrate-stimulated) oxygen consumption rates. Frontal cortex mitochondria from WT and ClockΔ19 mice were also used for protein and gene assays to compare mitochondrial subunit expression across oxidative phosphorylation (OXPHOS) complexes. Mitochondrial protein lysates were run on a Western blot and blotted using an antibody cocktail of against critical subunits of each of the 5 OXPHOS complexes (Abcam ab110413). qPCR was run for mitochondrial genes as previously described. Results: Human postmortem studies conducted in the PFC of patients with BD have shown a decrease in mitochondrial complex I function and expression. Our data shows a similar decrease in complex I-driven respiratory rate as determined by the addition of glutamate-malate and ADP to synaptosomal mitochondria from the PFC of ClockΔ19 mice. Additionally we demonstrate alterations to protein and gene levels specific to complex I and its downstream targets. Conclusion: These results show a decrease in mitochondrial expression and respiratory function that can be attributed to alterations in complex I in the presence of a dominant negative CLOCK protein. As the “entry enzyme” of cellular respiration complex I integrity has significant implications for ATP production management of reactive oxygen species (ROS) levels and maintenance of the NAD-NADH ratio. Significance: Through these preliminary studies we demonstrate that the ClockΔ19 mouse as a model for bipolar mania recapitulates the mitochondrial alterations found in human postmortem tissue and will serve as a model for future studies investigating the direct links between circadian clock machinery cellular metabolism and mitochondrial respiration.

First Author: Jared Kopelman (Graduate)	Poster Session: pm
Presenting Author: Jared Kopelman (Graduate)	Location: 47
Mentor/Lab: Ahmari	Category: Psychiatry
Department: Psychiatry	

Title: The Role of Candidate Gene Slc1a1 in OCD-relevant Behaviors in Mice

Summary: Obsessive Compulsive Disorder (OCD) is a debilitating psychiatric disorder characterized by intrusive obsessive thoughts and compulsive behaviors. Many studies have indicated that genetics play a significant role in the development of OCD. Here we investigate the role of Slc1a1 a gene associated with OCD in humans on behavior in mice.

Abstract: Obsessive Compulsive Disorder (OCD) is a debilitating psychiatric disorder characterized by intrusive obsessive thoughts and compulsive behaviors. The cause of OCD is unknown but human imaging studies have consistently shown hyperactivation of corticostriatal circuit nodes in patients with OCD. In addition twin and family studies show a significant role for genetics in its etiology with multiple studies identifying association of polymorphisms in the gene SLC1A1 with OCD. The most common of these OCD-associated polymorphisms increases expression of the encoded protein– the neuronal glutamate transporter excitatory amino acid transporter-3 (EAAT3). This protein is expressed in OCD-relevant corticostriatal circuits where it plays several roles including modulating the activation of peri-synaptic glutamate receptors. The OCD-linked allele is associated with increased SLC1A1 expression in lymphoblastoid cells human postmortem brain a luciferase reporter assay and transfected HEK cells where there is also a functional increase in EAAT3 protein activity as evidenced by increased glutamate uptake. There is also increased EAAT3 protein expression in striatum of Sapap3-knockout (KO) mice a model of OCD-like behavior. To directly test the effect of manipulations of EAAT3 levels on OCD-like behavior we used the Flexible Accelerated STOP Tetracycline Operator-knockin (FAST) system which combines cre flippase and tTA technology to create mice with either ablated or increased EAAT3 expression. Slc1a1-STOP knock-in mice that have ablated EAAT3 protein expression and function show blunted responses to pharmacologically-induced repetitive behavior. These mice have attenuated increases in stereotypy and hyperlocomotion in response to amphetamine and attenuated grooming increases in response to a dopamine D1 receptor agonist (Zike et al PNAS in press). Slc1a1-Overexpressing (OE) mice were created by breeding Slc1a1-tetO mice with CamKII-tTA mice. Slc1a1-OE mice show increased striatal EAAT3 expression that can be normalized by the administration of doxycycline allowing us to study the effects of temporally-specific EAAT3 overexpression. Here we present data from the initial behavioral characterization of Slc1a1-OE mice including OCD-relevant behaviors such as perseverative grooming pre-pulse inhibition and anxiety-like behavior.

First Author: Sean Piantadosi (Graduate)	Poster Session: pm
Presenting Author: Sean Piantadosi (Graduate)	Location: 48
Mentor/Lab: Ahmari	Category: Psychiatry
Department: Psychiatry	

Title: Using in vivo microscopy to assess the role of striatal medium spiny neurons in compulsive behavior and response to pharmacological treatment

Summary: The therapeutic mechanisms of the leading drug treatment for obsessive compulsive disorder are poorly understood. In this study brain activity was measured in a region called the striatum in a leading genetic mouse model of OCD using miniature microscopes that can visualize individual brain cells. This revealed that brain cells were overactive in the OCD model at baseline and that treatment with an effective OCD drug therapy normalized this activity.

Abstract: Study: Perseverative thoughts and actions are hallmark symptoms of Obsessive Compulsive Disorder (OCD) and are often present in other severe neuropsychiatric illnesses including autism and schizophrenia. Aberrant activity in cortico-striatal circuitry has been linked to compulsive behavior in both correlative studies in humans and causal studies in rodents. Using head-mounted mini-microscopes for in vivo calcium imaging (Inscopix) we sought to determine the role of medium spiny neurons (the principal striatal cell type) in mediating compulsive behavior in mice with a highly penetrant compulsive grooming phenotype (Sapap3-KO mice). We have also investigated how the first-line OCD pharmacotherapy the selective serotonin reuptake inhibitor fluoxetine alters striatal activity patterns. Methods: Sapap3 knockout (KO) mice which have both a hyperactive striatum and compulsive OCD-like grooming phenotype were injected with AAV-GCaMP6m and implanted with a GRIN lens in the centromedial striatum (CMS) to visualize striatal calcium activity during spontaneous grooming behavior. All mice received 7 days of treatment with the SRI fluoxetine (5 mg/kg) and underwent imaging and grooming assessments on days 3 5 and 7 of treatment. Results: At baseline Sapap3-KO mice displayed elevated grooming behavior and increased calcium activity during grooming relative to WT mice. This increase in calcium activity may stem from a strong increase in striatal activity at the onset of grooming events a phenomenon that was not observed in WT mice. Further activity of D1-MSNs is elevated at a trend level in Sapap3-KO mice suggesting an increase in direct pathway drive. Treatment with the SRI fluoxetine reduced observed calcium activity in all striatal cells with a rapid (3 day) time-course. Ex vivo data suggest that fluoxetine may be modulating the activity of striatal fast spiking interneurons (FSIs) in order to normalize striatal activity. Ongoing work is further dissecting striatal patterns that may contribute to compulsive behavior and its treatment. Conclusion: Hyperactivity of the striatum and compulsive grooming behavior can be reversed with successful SRI treatment in a valid mouse model of OCD-like behaviors. Significance: Understanding cell-type specific effects of successful and unsuccessful SRI treatment may help us develop treatments for patients with improved efficacy and fewer side effects.

First Author: Elizabeth Manning (Postdoctoral)	Poster Session: pm
Presenting Author: Elizabeth Manning (Postdoctoral)	Location: 49
Mentor/Lab: Ahmari	Category: Psychiatry
Department: Psychiatry	

Title: Impairments in cognitive flexibility relevant to OCD and accompanying alterations in cortico-striatal activity in SAPAP3 knockout mice

Summary: Patients with obsessive compulsive disorder (OCD) have trouble flexibly adapting their behavior. A leading genetic OCD mouse model was examined in a flexible decision making task. This revealed that approximately half of the OCD mice failed the flexible decision making task and the half that were successful showed activation of specific brain circuits that aren't activated in normal mice during flexible decision making.

Abstract: Background: Functional imaging studies have strongly implicated cortico-striatal circuit dysfunction in the pathophysiology of obsessive compulsive disorder (OCD). However the mechanisms by which this dysfunction gives rise to OCD symptoms are unclear with hyperactivity typically observed at baseline and during symptom provocation and hypoactivity typically observed during cognitive testing. Studies in preclinical rodent models provide a unique opportunity to investigate this discrepancy. To date transgenic mouse models have provided substantial insight about striatal dysfunction underlying OCD-relevant compulsive grooming. In contrast there have been no studies examining the neural mechanisms underlying cognitive impairment in OCD-relevant mouse models. Methods: SAPAP3 knockout mice (KOs)–a leading transgenic OCD model– and wild-type (WT) littermate controls were tested in an operant reversal learning paradigm to assess cognitive flexibility. Cortical and striatal activation associated with training on day 1 of reversal learning was assessed in a separate cohort of mice via quantitative cFos expression in 10 regions of interest (ROIs). Interactions between regional cFos expression genotype and reversal performance (correct vs incorrect lever presses) were assessed using linear regression analysis. Results: SAPAP3 KOs were significantly impaired in reversal learning (p<0.001) with ~40% of mutant mice (n=13/29) failing to acquire a reversed contingency (criteria: <20 active lever presses per day across 5 days of reversal training). Reversal learning impairment was unrelated to the severity of compulsive grooming observed in SAPAP3 KOs. Impaired reversal learning was also observed in female SAPAP3 KOs (n=9 WT 8 KO; 4 KOs failed reversal and 4 KOs acquired the task; training day x active lever press interaction p=0.008). Analysis of reversal learning-related cFos expression revealed genotype differences in the association between activity in the prelimbic prefrontal cortex (PrPFC) and nucleus accumbens shell (NAcS) and reversal performance (response ~ Genotype x ROI cFos x lever press type p<0.005). Both regions appeared to show compensatory neural activity in SAPAP3 KOs which was associated with improved acquisition of correct lever pressing following reversal. No such relationship was observed in WT mice. Conclusions: Our studies are among the first to describe neurocognitive impairments in a transgenic OCD mouse model. These findings implicate compensatory neural activity in the PrPFC-NAcS circuitry in successful reversal learning in SAPAP3 KO mice in line with recent studies demonstrating that stronger functional connectivity in cortico-striatal circuits is associated with intact cognition in OCD patients (Vaghi et al. 2017). Ongoing studies using in vivo microscopy to measure neural activity in SAPAP3 KOs during reversal learning are directly testing this hypothesis. Our results also highlight the utility of using OCD-relevant cognitive paradigms in preclinical mouse models to gain mechanistic insight regarding the role of cortico-striatal circuit dysfunction in OCD.

First Author: James Hyde (Postdoctoral)	Poster Session: pm
Presenting Author: James Hyde (Postdoctoral)	Location: 50
Mentor/Lab: Susanne Ahmari	Category: Psychiatry
Department: Psychiatry	

Title: In vivo calcium imaging of SKF38393 induced perseverative grooming in awake behaving mice

Summary: This study examines the neural activity in the ventral medial striatum during repetitive grooming in a pharmacological mouse model of OCD. We separated grooming behaviors into multiple types of grooming and showed that pharmacologically induced repetitive grooming selectively affects facial grooming rather than body grooming. We also showed that neural activity decreased during facial grooming during both pharmacologically induced repetitive grooming and normal grooming.

Abstract: Obsessive compulsive disorder (OCD) is characterized by intrusive obsessive thoughts and abnormal repetitive behaviors. Studies of several independent mouse models of OCD-like behavior suggest that perseverative grooming in mice is related to compulsive behaviors seen in OCD. Understanding the mechanisms leading to the development of abnormal grooming is therefore relevant to OCD pathophysiology. However the changes in cellular activity that are correlated with the development of perseverative grooming are unknown. Using miniaturized head-mounted microscopes and calcium imaging we therefore examined changes in cellular activity in the ventromedial striatum (VMS) during pharmacologically- induced perseverative grooming behavior. Drd1a-tdTomato mice were injected with the genetically encoded calcium indicator AAV9.hsyn.GCaMP6m and implanted with a microendoscope (6.1mm x 0.5mm GRIN lens) in VMS. Four weeks after virus injection mice were fitted with a microscope baseplate. Upon recovery behavioral experiments were performed. Using a cross-over within subjects experimental design mice were treated with either vehicle or the D1 agonist SKF38393 to induce perseverative grooming. Both behavior and calcium signaling were monitored continuously for 10 minutes prior to injection and 30 minutes post injection. Calcium data were extracted from processed videos to analyze event frequency and time-locked activity; both PCA/ICA and CNMF algorithms were used. As expected grooming activity increased after SKF38393 injection in VMS implanted mice. However we also found that SKF38393 selectively induces increased grooming activity only during the facial grooming steps of a stereotyped grooming chain. In vivo microendoscopy demonstrated that average calcium event rates decreased during facial grooming regardless of SKF38393 or saline treatment. However event rates selectively increased during non grooming and body grooming periods in SKF treated mice. Event rates during saline control experiments showed no differences between grooming and non-grooming time periods. These results suggest selective changes in striatal firing patterns as well as changes to initiation transition and cessation of grooming behavior after SKF38393 treatment. Ongoing analysis is delineating the precise relationship between changes in network level activity and bouts of perseverative grooming and determining whether the SKF-induced differences in event rates during non-grooming time periods are related to transitions into and out of grooming bouts.

First Author: Victoria Corbit (Graduate)	Poster Session: pm
Presenting Author: Victoria Corbit (Graduate)	Location: 51
Mentor/Lab: Susanne Ahmari Aryn Gittis	Category: Psychiatry
Department: Neurobiology	

Title: Dysregulation of specific cortical inputs to central striatum in the OCD-relevant Sapap3-KO mouse model

Summary: Imaging studies in Obsessive-Compulsive Disorder patients identify brain circuits showing dysregulated activity but the specific cell types and circuits are more easily studied using animal models. This work identifies a circuit imbalance such that a movement region has a strong influence on behavioral selection in an OCD-relevant mouse model whereas a cognitive flexibility region controls behavioral selection in healthy mice. These two regions are the best targets for brain stimulation treatment for OCD and these data provide evidence for how they may be important in OCD.

Abstract: Obsessive-Compulsive Disorder (OCD) is defined by the inability to suppress obsessive thoughts and compulsive behaviors. The exact neuronal mechanisms underlying these symptoms are unclear however hyperactivity in corticostriatal circuits is consistently observed in OCD patients. The Sapap3-KO OCD mouse model shows dysfunction in homologous corticostriatal circuits particularly those involving central striatum (CS) and lateral orbitofrontal cortex (LOFC). Specifically striatal fast-spiking interneurons (FSIs) are implicated because their aberrant regulatory influence over striatal output neurons (MSNs) is thought to play a role in abnormal behavioral selection. Both FSIs and MSNs are driven primarily by cortical inputs so investigating how specific cortical projections influence these CS cells is essential to understanding how these microcircuits contribute to compulsive behaviors. While LOFC is identified as an affected region in OCD patients CS is largely understudied. We first sought to characterize the major cortical afferents to CS. Using retrograde tracing we observed the well-known LOFC projections to CS but also unexpectedly identified a second major projection source M2. M2 has been suggested to be homologous to supplementary motor regions in humans and subregions of M2 in rodent are involved in the motor preparation while LOFC is important for cognitive flexibility. Thus both major inputs to CS are involved with behavioral selection and dysregulation in these inputs may play a role in the aberrant compulsive-like behaviors displayed by this OCD-relevant mouse model. To determine how specific cortical projections regulate microcircuits in CS we injected channelrhodopsin2 (ChR2) into cortex and recorded optogenetically-evoked excitatory post-synaptic currents (EPSCs) using acute slice physiology. LOFC-evoked EPSCs onto MSNs were weaker in KO mice relative to WTs while LOFC inputs to FSIs were unchanged. The ratio of EPSC amplitudes confirmed that LOFC input to FSIs is increased relative to nearby MSNs suggesting LOFC-evoked feedforward inhibition is stronger in Sapap3-KOs. In contrast we found that M2-evoked EPSCs were increased onto both MSNs and FSIs in the CS of Sapap3-KOs relative to WTs indicating a general increase in CS drive from M2. These data suggest that M2 rather than LOFC may be the primary source of cortical control of CS in Sapap3-KO mice potentially causing aberrant grooming behavior. This finding has particular relevance to OCD treatments as LOFC and supplementary motor area have been identified as the best targets for repetitive transcranial magnetic stimulation treatment in OCD patients. These results bring new focus to the role of supplementary motor cortical regions in the pathology of OCD and provide a foundation for future studies on M2 in compulsive behaviors in rodent models. Moreover increased relative drive to CS FSIs suggests that interneuron dysfunction may play a role in abnormal behavioral selection and initiation mechanisms relevant to OCD. Taken together these results reveal corticostriatal abnormalities that may cause compulsive behaviors in OCD.

First Author: Joanne C. Beer (Graduate)	Poster Session: pm
Presenting Author: Joanne C. Beer (Graduate)	Location: 52
Mentor/Lab: Howard J. Aizenstein Stewart J. Anderson and Robert T. Krafty	Category: Psychiatry
Department: Biostatistics Psychiatry	

Title: Predicting Social Responsiveness Scale scores of autism spectrum disorder patients from resting state fMRI data using structured sparse penalized regression

Summary: Can resting state functional connectivity predict Social Responsiveness Score in autism patients? We propose a novel penalized regression estimator that is informed by spatial and functional relationships between neuroimage voxels. We apply the estimator to resting state fMRI data from the Autism Brain Imaging Data Exchange (ABIDE) Preprocessed dataset in order to pinpoint the cortical brain regions whose functional connectivity with a subcortical seed region best predicts Social Responsiveness Score.

Abstract: Penalized regression estimators such as lasso ridge regression or elastic net are often used in neuroimaging-based prediction models. These estimators yield unique solutions when data is high dimensional (i.e. when there are more predictors than subjects) by imposing optimization constraints that result in global sparsity or shrinkage of estimated coefficients. However often more is known about the relationships between predictors. For example when neuroimage voxels are used as predictors we might expect neighboring voxels to be similar to each other and therefore expect estimated coefficients to exhibit some degree of spatial smoothness. Additionally we might expect related voxels such as those residing in the same functional networks or anatomical regions to be selected or shrunk to zero as a group. We propose incorporating information about spatial and functional relatedness of voxels into the optimization constraints by using a fused sparse group lasso estimator. Lasso fused lasso group lasso and sparse group lasso are special cases. Simulation studies demonstrate conditions under which the fused sparse group lasso penalty yields better predictions than the lasso fused lasso group lasso or sparse group lasso penalties alone. We apply the fused sparse group lasso estimator to resting state fMRI data from the Autism Brain Imaging Data Exchange (ABIDE) Preprocessed dataset in order to pinpoint the cortical brain regions whose functional connectivity with a subcortical seed region best predicts Social Responsiveness Scale score.

First Author: Felipe Gomes (Postdoctoral)	Poster Session: pm
Presenting Author: Felipe Gomes (Postdoctoral)	Location: 53
Mentor/Lab: Anthony Grace	Category: Psychiatry
Department: Neuroscience Psychiatry and Psychology	

Title: The ability of stress during adolescence or adulthood to produce schizophrenia-like pathophysiology is dependent on the state of the critical period

Summary: Timing of the stress is a critical determinant of the pathophysiology that is present in the adult. While adolescent stress could led to changes that recapitulates schizophrenia adult stress induced changes observed in depression. Re-opening the sensitive period in the adult restores vulnerability to stress-induced pathology resembling schizophrenia.

Abstract: Title: The ability of stress during adolescence or adulthood to produce schizophrenia-like pathophysiology is dependent on the state of the critical period Felipe V. Gomes Xiyu Zhu Anthony A. Grace Departments of Neuroscience Psychiatry and Psychology University of Pittsburgh Background: Unregulated stress exposure occurring during the sensitive period of development leads to the emergence of circuit deficits consistent with schizophrenia in the adult. If accurate one would predict that re-opening the sensitive period in the adult could make it susceptible to a similar disruption. Methods: Male rats were submitted to a combination of footshock (FS) and restraint stress (RS) during adolescence (PD31-40) or adulthood (PD65-74). The activity of dopamine (DA) neurons in the ventral tegmental area (VTA) and the pyramidal in the ventral hippocampus (vHipp) were evaluated 1-2 or 5-6 weeks post-stress. We also evaluate if the administration of valproic acid (VPA; 300 mg/kg) which is known to re-instate the critical period in adults would recreate an adolescent phenotype of susceptibility to stress. Results: Our data suggest that as indicated by the increased VTA DA neuron population activity. The adolescent stress increased VTA DA population activity 1-2 and 5-6 weeks post-stress suggesting that adolescent stress induced both short- and long-term schizophrenia-like changes in the VTA DA system. These changes seem to be driven by an increased vHipp activity. On the contrary adult stress produced short-term depression-like changes as indicated by the decreased DA neuron population activity in the VTA which failed to persist after 5-6 weeks. Interestingly VPA treatment altered the impact of adult stress. When rats were treated with VPA FS+RS increased VTA DA population activity similar to that observed with adolescent stress. Conclusion: Timing of the stress is a critical determinant of the pathophysiology that is present in the adult. While adolescent stress could led to changes that recapitulates the MAM model of schizophrenia adult stress induced changes observed in animal models of depression. Re-opening the sensitive period in the adult restores vulnerability to stress-induced pathology resembling schizophrenia. Financial support: MH57440

First Author: Emily Parker (Graduate)	Poster Session: pm
Presenting Author: Emily Parker (Graduate)	Location: 54
Mentor/Lab: Sweet	Category: Psychiatry
Department: Psychiatry	

Title: Synaptic remodeling of small dendritic spines over adolescent auditory cortex development

Summary: Recently our group discovered that in schizophrenia in the primary auditory cortex (A1) the loss of dendritic spines important synaptic structures on excitatory neurons is driven by the selective reduction of the smallest spines and that the protein CaVβ4 could play a role in this loss. We hypothesized that like in schizophrenia spine loss in A1 is selective for the smallest dendritic spines during synaptic remodeling of excitatory circuits in normal mouse adolescent development and that CaVβ4 plays a role in this process. We found that the smallest spines are indeed selectively lost over mouse A1 adolescent development but could not confirm if CaVβ4 drives this loss as our data indicate that CaVβ4 levels modestly decrease and that levels of CaVβ4 were not significantly associated with reduced number of small spines over mouse A1 adolescent development.

Abstract: *This poster abstract was submitted to accepted and will be presented at SfN 2017 Dendritic spines are motile postsynaptic structures at excitatory synapses. Following synaptic remodeling of excitatory circuits during adolescence in normal development dendritic spine density is reduced in cortical areas including primary auditory cortex (A1) in adulthood. Excess synaptic remodeling during adolescence is thought to occur in schizophrenia (Sz) resulting in excessive loss of dendritic spines. Reduced dendritic spine density has been observed in multiple brain regions in Sz in adulthood including in A1. We recently reported that the dendritic spine density reduction in Sz in A1 is limited to dendritic spines of smaller volumes which are presumed to be predominantly transient. Further we found that increased levels of a peptide shared among CaVβ isoforms was associated with reduced density of small but not large dendritic spines in A1. Overexpressing CACNB4 which encodes CaVβ4 led to reduced density of small dendritic spines in primary neuronal culture. For the current study we hypothesized that previous observations of reduced A1 dendritic spine density during adolescent development is driven by and selective for small dendritic spine loss. We measured dendritic spine density over A1 adolescent development using stereological and quantitative confocal fluorescence microscopy techniques and found that mean density of small dendritic spines was significantly reduced in adult (P84) as compared to early adolescent (P28) mouse A1 (p<.001). The density of large dendritic spines was not altered. These findings suggest that the smallest and likely transient dendritic spines are targeted during synaptic remodeling in A1 during adolescent development. We will report findings from experiments that characterize CaVβ levels over normal A1 mouse development to determine if elevated CaVβ4 levels are associated with dendritic spine density reduction during adolescent development in mouse A1.

First Author: Jillian Weeks (Graduate)	Poster Session: pm
Presenting Author: Jillian Weeks (Graduate)	Location: 55
Mentor/Lab: Sved	Category: Psychiatry
Department: Neuroscience	

Title: Nicotine reinforcement is not increased in the MAM rodent model of schizophrenia

Summary: Individuals with schizophrenia smoke at a rate 4 to 5 times higher than the general population and with greater frequency and intensity but the mechanism behind this is unknown. This experiment used an animal model of schizophrenia to determine if increased reward from nicotine the primary psychoactive component of tobacco drives this increase in smoking.

Abstract: Despite progress in reducing smoking over the past several decades up to 80% of individuals with schizophrenia (SCZ) continue to smoke. SCZ patients also smoke more intensely and with greater frequency contributing to a disproportionately negative impact on health. However no clear mechanistic connection between SCZ and smoking has been established. One hypothesis underlying the behavior is that SCZ brain pathophysiology confers an increased propensity to take nicotine (NIC) the primary psychoactive component of cigarette smoke. We sought to characterize NIC reinforcement as measured through a self-administration paradigm in a neurodevelopmental rat model of SCZ. Pregnant dams were treated with either methylazoxymethanol acetate (MAM; 1 mg/kg i.p.) or saline (CTL) on gestational day 17. Adult male and female offspring were allowed to self-administer NIC across a range of doses (0 - 60 micrograms/kg/infusion 7 days/dose) paired with neutral cue (CS) or reinforcing visual stimulus (VS) in daily 1 hr sessions. MAM and control rats did not differ in infusions + CS earned at any NIC dose (e.g. 15 microgram/kg/infusion dose females; MAM n = 22 mean = 9.8 ± 1.2 infusions; CTL n = 18 mean = 9.9 ± 0.9 infusions). MAM rats earned fewer infusions of NIC paired with VS at all doses tested (e.g. 30 microgram/kg/infusion dose males; MAM n = 9 mean = 17.2 ± 1.4 infusions; CTL n = 10 mean = 21.9 ± 1.3 infusions) but also responded less for VS alone. This suggests that VS may be less reinforcing to MAM animals which may in turn reduce the relative magnitude of NIC enhancement of VS reinforcement. To capture patterns of responding across an extended period rats in a separate experiment were allowed to self-administer NIC + CS for 23-hr sessions. No differences in NIC-taking between MAM and CTL rats were observed in 23-hr sessions. Overall GD17 MAM did not produce an increase in NIC self-administration in male or female rats which suggests that SCZ pathophysiology as modeled in these animals does not elevate NIC intake due to increased NIC reinforcement.

First Author: Darius Becker-Krail (Graduate)	Poster Session: pm
Presenting Author: Darius Becker-Krail (Graduate)	Location: 56
Mentor/Lab: Colleen McClung	Category: Psychiatry
Department: Psychiatry (TNP)	

Title: Circadian transcription factor NPAS2 and metabolic redox sensor SIRT1 interact in the mouse nucleus accumbens (NAc) to regulate cocaine reward-related behavior

Summary: Cocaine's effects on the metabolic state of the cell may feed into the circadian molecular clock and in turn alter reward regulation.

Abstract: Cocaine addiction is a widely prevalent substance use disorder in the United States. With a lack of successful therapeutic options it is important to investigate the cellular and molecular level changes following cocaine use and how these changes establish and/or reinforce addiction. As its mechanism of action cocaine increases mesolimbic dopaminergic signaling via inhibition of dopamine transporter. This increased activity is energy taxing for the cell and can cause both severe oxidative stress and altered mitochondrial function. Interestingly metabolic changes associated with cocaine use may directly regulate the circadian molecular clock and its output genes through associated metabolic redox sensors. More specifically the circadian transcription factors CLOCK/NPAS2 and the NAD+ dependent deacetylase SIRT1 have all been shown to directly respond to changes in levels of the mitochondrial coenzyme NAD+. Previous work in the lab has shown CLOCK and NPAS2 differentially regulate cocaine reward; e.g. mutations in the Clock gene increase cocaine preference and self-administration while mutations in Npas2 yields an opposite phenotype. Moreover our data suggest NPAS2 regulates reward through its enriched expression in the nucleus accumbens (NAc). Interestingly SIRT1 modulators have also been shown to regulate cocaine preference in that SIRT1 agonists increase cocaine preference and vice versa. In addition to NPAS2 and SIRT1 modulation altering cocaine reward chronic cocaine exposure has been shown to preferentially alter expression of these proteins in the NAc. Given these observations we investigated how changes in cellular metabolic state may feed into the circadian molecular clock and alter regulation of cocaine reward and whether an interaction between NPAS2 and SIRT1 in the NAc mediates this regulation. Through co-immunoprecipitation studies our preliminary findings suggest that NPAS2 and SIRT1 do interact in a shared complex in the NAc and chronic cocaine may alter this interaction. Furthermore utilizing high-performance liquid chromatography to assess NAD+ concentration we observed a diurnal variation of NAD+ levels in the striatum that is disrupted following chronic cocaine exposure. Finally in mice with a NAc specific viral-mediated knock-down of NPAS2 we determined NPAS2 to be necessary for the increase in cocaine preference seen with a SIRT1 agonist. Ultimately our findings highlight a mechanism by which chronic cocaine’s metabolic changes can directly alter circadian molecular clock function and how this interaction mediated by NPAS2 and SIRT1 may afford regulation of cocaine reward-related behavior.

First Author: Lauren DePoy (Postdoctoral)	Poster Session: pm
Presenting Author: Lauren DePoy (Postdoctoral)	Location: 57
Mentor/Lab: Colleen McClung PhD	Category: Psychiatry
Department: Psychiatry	
Title: Npas2 knockout increases intravenous cocaine self-administration	

Summary: Substance use is associated with changes in sleep/wake cycles and circadian rhythms and circadian genes appear to play an important role in regulating reward. Here a mutation in one gene Npas2 increases cocaine intake and motivation in a mouse model of drug taking. By understanding how circadian genes regulate reward we can develop novel treatments for substance dependence.

Abstract: The development of substance dependence is associated with disruptions in circadian rhythms and circadian genes. In mice a dominant negative mutation in circadian locomotor output kaput (CLOCK) increases both cocaine reward and self-administration. However the role of its homologue neuronal PAS domain protein 2 (NPAS2) in cocaine self-administration remains unclear despite Npas2 knockout contrastingly decreasing cocaine reward. We performed intravenous cocaine self-administration using male and female mice with a mutation in Npas2. Mice first acquired an operant response for food and then were implanted with an indwelling jugular catheter. After recovery mice acquired cocaine self-administration and then dose-response testing was conducted both at a fixed ratio and progressive ratio schedule. Npas2 knockout did not impact acquisition of a food response however it did accelerate acquisition of a cocaine-reinforced response as well as increase the total number of infusions earned. Furthermore Npas2 knockout increased the reinforcing and motivational properties of cocaine as evidenced by an upward shift in dose-response curve and an increase in breakpoint ratio respectively. Overall Npas2 knockout increases cocaine intake propensity to self-administer cocaine as well as the reinforcing and motivational properties of cocaine in mice across sex. This divergence from decreased cocaine reward seen in Npas2 knockout mice is likely due to the volitional control over drug intake during self-administration compared to conditioned place preference. Further research is required to understand the differences between NPAS2 regulation of cocaine reward and drug consumption.

First Author: Megan Bertholomey (Postdoctoral)	Poster Session: pm
Presenting Author: Megan Bertholomey (Postdoctoral)	Location: 58
Mentor/Lab: Torregrossa	Category: Psychiatry
Department: Psychiatry	

Title: KETAMINE REDUCES YOHIMBINE+CUE-INDUCED REINSTATEMENT OF ETHANOL SEEKING AND DEPRESSIVE-LIKE BEHAVIOR IN FEMALE RATS.

Summary: Females represent a vulnerable population with respect to stress-related disorders like depression and alcoholism indicating better treatment for women is needed. Low doses of ketamine have been shown to produce antidepressant and stress-blocking effects in male humans and animals. We show that ketamine not only blocks depression-like behavior in female rats but it also blocks stress-related alcohol drinking and seeking.

Abstract: Alcohol use and major depressive disorder are frequently comorbid with individuals diagnosed with a substance use disorder being nearly three times as likely to have major depression. Poor treatment responses are found for both disorders and are further complicated when they co-occur underscoring the need for better therapies. One promising candidate is ketamine which has been shown to have rapid and long-lasting effects in individuals with treatment-resistant depression and in rodent stress models. Ketamine has also been shown to reduce ethanol drinking in male Sardinian alcohol-preferring rats. However though women are more likely to have this comorbidity few studies have examined sex-specific effects of ketamine on depressive symptoms and none have done so for alcohol drinking or seeking. Therefore the goal of the present experiment was to determine both acute and long-term effects of ketamine on both alcohol-motivated and depressive-like behaviors in female rats. Rats were injected with an antidepressant dose of ketamine (10mg/kg) an anesthetic dose of ketamine (90mg/kg) or saline at postnatal day p27 and were trained to self-administer a 10% ethanol+0.1% saccharin solution in young adulthood beginning around p70. Though adolescent pretreatment with ketamine did not alter ethanol self-administration acute ketamine (10mg/kg) treatment robustly reduced cue+yohimbine-induced reinstatement of ethanol seeking which tended to last up to 3 weeks post-treatment in subsequent reinstatement tests in rats that had received the same antidepressant dose of ketamine prepubertally. This suggests that antidepressant doses of ketamine may cause neuroadaptive changes during development that increase the sensitivity to the protective effects of acute ketamine exposure in mitigating stress-related behaviors in adulthood. During subsequent forced swim testing ketamine produced significant decreases in immobility suggesting an antidepressant effect. Thus acute ketamine treatment reduces both alcohol-motivated and depressive-like behavior under stressful conditions in female rats. These data confirm the antidepressant effects of ketamine in female rats that have been previously shown in males but also demonstrates that ketamine may be an effective treatment for stress-induced alcohol seeking. Ongoing studies are aimed at identifying the neural mechanisms underlying these effects and expanding these findings to determine the effects of ketamine on stress-related alcohol or saccharin drinking in both male and female rats.

First Author: Kelly Barko (Faculty)	Poster Session: pm
Presenting Author: Kelly Barko (Faculty)	Location: 59
Mentor/Lab: Logan/Dr. Ryan Logan	Category: Psychiatry
Department: Translational Neuroscience Program Department of Psychiatry	

Title: Circadian Rhythms and Opiates: Role of the Circadian Transcription Factor NPAS2 to Regulate Morphine Conditioned Reward

Summary: Acute and chronic substance use such as morphine can cause disruptions in circadian rhythm. The molecular mechanism behind drug-related behavior with respect to circadian rhythm remains uncertain. Thus our current research focuses on exploring a putative cell-specific type mechanism post morphine administration.

Abstract: Background There is evidence supporting substance use such as psychostimulants or opiates can cause disruption in endogenous circadian rhythms. However the molecular mechanism behind the pathophysiology of mood and addiction disorders with respect to circadian rhythm remains uncertain. Located within the striatum of the mammalian forebrain is the reward center of the brain known as the nucleus accumbens (NAc). NPAS2 an integral basic helix-loop-helix (bHLH)-PAS transcription factor of the molecular clock found throughout the NAc is expressed in medium spiny neurons (MSNs) that contain dopamine subtype receptors 1 (D1+) or 2 (D2+). According to our previous studies an increase in NPAS2 expression was observed when D1+ MSNs were activated post psychostimulant administration. Thus our current research focuses on manipulating NPAS2 within the NAc of D1+ or D2+ MSNs to explore a potential cell-type specific role in the behavioral response to morphine. Methods Wild-type and NPAS2-bHLH-deficient male and female mice underwent unbiased morphine CPP. We also designed a Cre-inducible shRNA virus (AAV2) to knockdown Npas2 (or Scramble control) specifically in D1+ or D2+ MSNs by stereotaxic injection into the NAc of D1-Cre or D2-Cre mice. The NAc of extracted brains were punched and used for molecular assays including qPCR Western blots and protein IP. Results Acute and chronic morphine administration altered the expression of NPAS2 in the NAc. Wild-type male and female mice showed an expected preference for the morphine-paired side. NPAS2 KO male mice displayed a significantly attenuated development of morphine CPP. Cell-type specific knockdown of Npas2 in D1+ MSNs of the NAc also significantly attenuated morphine CPP in male mice with moderate effects in D2+ MSNs. Concluding Statement Although a definitive singular cellular mechanism remains unclear we will continue to investigate the role(s) of NPAS2 within the NAc in relation to substance use and the circadian pathway.